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**Proceedings of the
40th Southern Pasture
and Forage Crop
Improvement
Conference**

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Held at Baton Rouge, Louisiana,
April 16-19, 1984

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Proceedings of the 40th Southern Pasture and Forage Crop Improvement Conference

Held at Baton Rouge, Louisiana,
April 16-19, 1984

Sponsored by the
Agricultural Experiment Stations of
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PREFACE

These Proceedings include most of the reports and papers presented at the 40th meeting of the Southern Pasture and Forage Crop Improvement Conference held April 16-19, 1984, at Baton Rouge, Louisiana, with Louisiana State University as host. On April 17 after welcoming remarks by Dr. H. R. Caffey, Chancellor of LSU, participants were introduced State by State. The research program of the Louisiana Agricultural Experiment Station for animals and crops was discussed as was the Cooperative Extension Service Program. In the afternoon, Research Information Exchange Groups (formerly Work Groups) met with many presentations which will be reported in subsequent pages. A banquet was held on the evening of April 17 with a speech by Floyd Kent, County Agent, on Louisiana Agriculture and Its People.

On April 18, the group toured "Florida Parishes" in southeast Louisiana. At the Southeast Research Station, Franklinton, the group visited the new Forage Quality Evaluation Laboratory and saw the research being done on limited-tillage. After lunch, the group toured the Heyward Green Farm where embryo transplants were demonstrated. At the Kirby Varnado farm, forages including silage utilization was seen on its Holstein herd which has a rolling average of 20,000 pounds of milk.

The Conference concluded with a General Session for papers and a General Business Meeting on the morning of April 19. Details of the Business Meeting and a List of Conference Registrants are at the end of these Proceedings.

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Carl S. Hoveland¹

INTRODUCTION

The historic discovery by Dr. Joe D. Robbins and co-workers (Robbins 1983) at the Russell Research Center in Athens, Georgia, during the 1970's that a fungal endophyte was associated with fescue toxicosis spawned a great deal of research in many states that has made it possible to obtain good animal performance on this well adapted grass. From this work has come a better understanding of the fungal endophyte and practical methods for its control. Although much has been learned, the toxic agent has eluded scientists, and the mode of action in the animal is still imperfectly understood.

The tall fescue-endophyte association is a fascinating story and has aroused great interest among both scientists and livestock producers. A number of workshops and seminars dealing entirely or partially with this problem have been held in States such as Georgia (Anon. 1983 a), Oregon (Anon. 1983 b), Arkansas (Anon. 1983 c), and Missouri (Anon. 1984) which resulted in published proceedings. Several popular reviews on this subject have also been published (Ball 1984, Ball and Hoveland 1983, Lacefield 1983).

This paper will not attempt to cover all the literature on this subject but rather to focus on our present knowledge of the fescue-endophyte association from current published and unpublished research. It is hoped that this overview will sketch the current status of research and some of the unsolved problems in fescue toxicosis or "summer syndrome".

The endophyte associated with tall fescue toxicosis was originally identified as Epichloe typhina (Bacon et al. 1977). However, further studies have reclassified it as Acremonium coenophialum (Morgan-Jones and Gams 1982).

ANIMAL PERFORMANCE

Grazing and feeding trials clearly show that cattle performance on tall fescue is greatly improved when Acremonium coenophialum levels are low as compared to the usual poor performance when tall fescue endophyte levels are high. In a 3-year Alabama grazing trial, beef brood cows lost weight on infected tall fescue while cows on low endophyte grass gained nearly 0.5 kg/day (Schmidt et al. 1983). Calf daily gain was 50% higher on low endophyte than on heavily infected pastures, reflecting the 80% higher milk production per cow on low

endophyte grass. Reproduction of animals grazing infected tall fescue is adversely affected, but research data is lacking on this subject.

Steer average daily gain (ADG) over four years in Alabama was 82% higher on tall fescue pasture with less than 5% endophyte than with 94% infected plants (Hoveland et al. 1984 b). At low endophyte levels, ADG's of 0.83 kg were obtained over the grazing season, indicating the high potential of tall fescue (Hoveland et al. 1983). Although this syndrome has often been called "summer syndrome" because of poor performance and appearance of animals in warm weather, ADG's of steers have been depressed as much or more during the cool and autumn winter months on endophyte-infected tall fescue (Hoveland et al. 1984 a). Visible symptoms of the syndrome increase with higher ambient temperature (Hemkin et al. 1981). In very short-term spring grazing trials in Alabama, any infestation level was harmful to steer ADG, decreasing an average of 0.09 kg for every 10% fungal infestation level (Stuedemann, et al. 1984). In another 3-year Alabama grazing trial at endophyte levels of 0, 28, and 90% the total season ADG of steers on KY 31 tall fescue pasture was 0.98, 0.80, and 0.64 kg, respectively (Pedersen et al. 1984 e). At this point, it is difficult to predict animal response to a particular level of endophyte infestation. Since endophyte levels increased from April to late June in Georgia, sampling time may be critical in assessing infection level (Belesky et al. 1984). Hay and seed of infected tall fescue also result in depressed gain when fed to steers (Schmidt et al. 1982).

Performance of dairy cattle on endophyte-infected fescue is also depressed (Hemken et al. 1984). Symptoms such as elevated body temperature, increased respiration rate, reduced feed intake, decreased milk yield, rough hair coat, and depressed serum prolactin level are similar to that observed in beef cattle. Milk persistence of cows grazing low endophyte tall fescue in Alabama was similar to that on annual ryegrass (C. C. King, Jr., unpublished data).

Visible symptoms of fescue toxicosis on endophyte-infected grass were most obvious with a higher rate of nitrogen fertilizer, but this had no effect on ADG (Stuedemann et al. 1984). Total rumen volatile fatty acid concentration was greatest in low endophyte and low nitrogen fertilized fescue and least on high endophyte grass (McHan et al. 1984). Nitrogen fertilization resulted in a higher proportion of short chain acids as compared to long chain acids in endophyte-infected tall fescue. The effects of the endophyte on forage digestibility, rumen metabolism, intake, and utilization by the animal are not well understood and need further research.

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A great deal of research has been done in recent years by Bacon and associates at the Russell Research Center, Athens, GA, on the life cycle, morphology, ultrastructure, and nutrition of the Acremonium coenophialum endophyte. Much of this research has been reported in several conference proceedings (Anon. 1983 a, 1983 b, 1984) and will be highlighted here.

Endophyte-infected tall fescue plants have no external symptoms and flower and set seed normally, resulting in a unique relationship between fungus and grass (Bacon and Hinton 1983). It is a seed-borne pathogen in which the fungus hyphae are located between the aleurone and endosperm layers but grow into the starchy endosperm layer. As the seedling develops, it requires 4 to 5 weeks for invasion of the fungus into leaf tissue where it does not penetrate the host cytoplasm but remains in the intercellular spaces. The greatest concentration of fungus is found in the leaf sheath, seeds, crowns, and stems with little or none in root tissue. Nutrient usage by the fungus from the host plant appears to be minimal. However, the fungus does cause the production of pyrrolizidine alkaloids and phytoalexins. Research is in progress to determine the role of these compounds in fescue toxicosis of animals.

A comprehensive paper by Siegel et al. (1984 a) in Kentucky reports on the incidence and dissemination of the tall fescue fungal endophyte. Pastures of Kentucky 31 tall fescue in Kentucky are heavily infested with the fungus. The widespread occurrence of the fungus can be traced to its origin. The high number of infected ecotypes from Europe, the high infestation level on the Suiter farm in Kentucky where Kentucky 31 was selected, and rapid acceptance by farmers and planting of current year's seed production probably account for the high incidence of the fungus in pastures. The high demand and planting of Kentucky 31 tall fescue throughout the current "fescue belt" during the late 1940's and early 1950's utilized highly infected seed as there was no opportunity for the fungus to die as it would be if stored for a year or more. Since tall fescue pastures generally were permanent, there was no opportunity to replant them with old seed. Storage of infected seed at 21°C generally results in total loss of endophyte viability after 7 to 11 months but no loss after 19 months storage at 6°C.

Spread of the endophyte appears to be via seed with no dissemination by pollen, wind, or rain. Mowing and trampling by animals also have no effect on dissemination except as seed may be scattered. One disturbing report concerns an apparent increase in percent infection of a tall fescue seed field in Alabama after four years (Pedersen 1984 b). This suggests that yearly monitoring of seed production fields will be necessary and continued research is

needed on this subject.

Funk et al. (1983) have found that infected ryegrass plants have enhanced insect resistance. This has not been reported in field-grown infected tall fescue. However, laboratory studies have shown reduced insect feeding on infected tall fescue in New Zealand (Siegel et al. 1984 a). Also, in pastures it is possible that endophyte-infected plants may have a selective advantage over endophyte-free plants as grazing animals select one in preference to the other. Further research will be needed to determine if a shift to endophyte-infected plants might occur over a long period of time. The effect of the endophyte on plant morphology and growth in a pasture has not been answered. However, Read et al. (1984) in Texas reported higher total dry matter production on endophyte infected than endophyte-free pastures. The finding that a fungal endophyte Balansia epichloe produced in culture a possible precursor of the plant auxin, indole acetic acid, 3-indole glycerol is also intriguing (Porter et al. 1977).

TOXINS ASSOCIATED WITH THE ENDOPHYTE

Pyrrolizidine alkaloids (N-formyl loline and N-acetyl loline) appear to be associated with the fungal endophyte (Jones, et al. 1983). Loline concentrations and endophyte levels increased from April to late June in Georgia pastures with highest concentrations at high nitrogen fertilization (Belesky, et al. 1984). However, the loline alkaloids have not been found in endophyte-infected perennial ryegrass causing the staggers syndrome in sheep (Siegel et al. 1984 a). Garner (Garner 1983) has reported that the cation fraction (non-alkaloid) contained the summer syndrome toxin, casting doubt upon alkaloids as a causal agent.

Researchers at Auburn University (N. D. Davis, P. A. Backman, E. M. Clark, and S. P. Schmidt, unpublished data) have isolated by mass spectrometry a mycotoxin present both in endophyte-infected tall fescue and in fungus grown on a media. The molecular weight of 429 is significant in that it is an odd number, thus having an odd number of N's, and so is in a different class of alkaloids than the lolines and perlolines. It may be in the class of tremogens found in perennial ryegrass in New Zealand. It is possible that it may be a paspalacine or paxaline alkaloid. Tested in chick embryos, it is extremely toxic. Thus, to date, the toxic agent responsible for fescue toxicosis has not been identified and demonstrated to produce fescue toxicosis in cattle. Some researchers also feel that phytoalexins should receive much more attention in the search for the toxic agent responsible for the syndrome.

DETECTION OF THE ENDOPHYTE

Since there are no visible symptoms of the endophyte in infected tall fescue, several methods have been devised to detect the endophyte (King et al. 1983). Microscopic examination of stained plant tissue is laborious and slow. An improved histochemical technique involving the leaf sheath has been described (Pedersen et al. 1984 b). This technique is appropriate for newly harvested seed or plant tissue samples but is not suitable for old seed since live and dead fungus cannot be distinguished.

The ELISA (enzyme-linked immunosorbent assay) involves the use of an antibody reaction to determine the presence of the fungus in freshly harvested seed on fresh plant samples. It only determines whether or not the fungus is present and does not determine if it is alive or dead.

A grow-out procedure is needed to determine the live fungus status of seed. When seedlings are grown to about two months of age, the staining test or ELISA can be used to analyze for the fungus. This test will detect if the fungus has died.

Several laboratories conduct diagnostic tests of seed and plant tissue. Auburn University has a Fescue Toxicity Diagnostic Center headed by Dr. Richard Shelby that analyzes samples on a fee basis. By February 20, 1984, this laboratory had analyzed 720 samples of plants and seed. Some results indicate the prevalence of fescue toxicity (courtesy Dr. R. Shelby):

<u>State</u>	<u>Total samples</u>	<u>Mean percent of samples infested</u>
Missouri	235	66%
Alabama	183	69
Mississippi	77	80
Oregon	39	10
Virginia	29	75
North Carolina	25	55
Oklahoma	17	80
Georgia	16	71
Ohio	16	58
Kansas	14	65

CONTROL OF THE ENDOPHYTE

Since surveys in several states indicate that most Kentucky 31 tall fescue pastures are infected with the endophyte, control methods have received considerable attention. In Kentucky, field application of the fungicides benomyl, triadimetan, thiabendazole, etaconazol, or imazalil did not control the endophyte in tall fescue (Siegal et al. 1984 b). In Alabama, application of triadimetan or propiconazole granules to infected tall fescue pastures reduced infestation levels from 70% to 36% and improved ADG of steers from 0.41 to 0.68 kg (Williams and Backman 1984). Unfortunately, endophyte levels reverted to pre-treatment levels on treated pastures by 7.5

months after treatment. Thus, field application of granular fungicides does not seem feasible because of the high cost of annual treatment. Chemical seed treatment with certain triazole fungicides has given good endophyte control in greenhouse trials and does not affect seed viability (Siegal et al. 1984 b, Williams and Backman 1984). However, endophyte control in fungicide-treated seed in the field has been erratic and apparently dependent on soil type.

On-farm storage for a year or a heat treatment are effective ways to eradicate the endophyte but seed germination will be reduced (Siegal et al. 1984 b, Williams and Backman 1984). Probably the best method of eradicating the endophyte is to replant pastures with tall fescue seed certified to be less than 5% infested. Seed certified to contain less than 5% endophyte are now available or will be in autumn of 1984 for a number of cultivars.

Replanting old pastures with endophyte-free seed should be most successful where all of the seed produced by the old fescue stand have become non-viable. Fescue seedlings were about eliminated after 18 months of being cropped with soybeans or fallowed in central Alabama (Pedersen et al. 1984 a). By preventing any established fescue from producing seed in the same year a new stand is to be planted, the remnant seed would be about 15 months old and few tall fescue seed would be expected to germinate.

In order to successfully reestablish low endophyte tall fescue pastures, the old sod must be completely destroyed. On land capable of being cropped, tillage and cropping with soybeans, corn, or grain sorghum would be desirable. However, much of the area currently in tall fescue is too hilly for tillage, resulting in severe soil erosion. In addition, tillage is expensive. Killing the sod with a herbicide such as Paraquat, Roundup, Poast, or Fusilade should be possible. Research in Tennessee (H. A. Fribourg, unpublished data) and in Georgia (C. S. Hoveland, unpublished data) suggest that endophyte-free tall fescue can be successfully drilled into an old infected sod that has been killed with a herbicide. The great potential improvement in animal performance on endophyte-free pastures, coupled with low cost and minimum soil erosion, should make this attractive for livestock producers.

SUMMARY

The association of the fungal endophyte Acremonium coenophialum with fescue toxicosis and the finding that low-endophyte tall fescue can greatly improve animal performance is a major breakthrough in grassland research. The unique relationship between fungus and grass where the endophyte appears to be transmitted only via infected seed offers an opportunity to reestablish low endophyte pastures that will

remain that way for a long time. Although the toxic agent responsible has not been identified and demonstrated to produce fescue toxicosis in cattle, some practical measures to detect the endophyte and control the problem have been devised. Further research is needed on plant-endophyte interactions, metabolism in the animal, and control measures. The cooperative research of many scientists on this problem has provided an opportunity to realize the full potential of tall fescue in improved livestock production.

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Although the phenomenon of poor establishment in warm-season grasses is well documented, few solutions to the problem have been proposed. We have studied factors associated with seedling establishment in *Eragrostis* and *Panicum* species for several years, and will summarize our results and those of others, as well as a few of our ideas, in an attempt to suggest methods to improve establishment of forages in general.

Many factors can lead to poor establishment, but in the more arid regions of the south and southwest, water availability is the most important factor. Inadequate and unpredictable rainfall, coupled with relatively high temperatures and low humidity, make the first several centimeters of the soil a hostile environment for seedling growth. Also, because forage grasses have relatively small seed, these seed cannot be "planted to moisture," as is the case with large seeded cereals or other field crops. Hence, forage grass seed are frequently planted in dry soil, and germinate only after a sizeable precipitation event. As soon as germination begins, each seedling is in a life or death struggle to maintain a favorable water balance until adventitious roots are extended several centimeters into the soil. One realistic goal for the physiologist and plant breeder is to modify the behavior of the developing seedling in order to shorten the period of time between germination and the penetration of adventitious roots to soil depths where a more dependable water supply should be available. For convenience, this discussion will be divided into sections based on stages of plant growth, ranging from the dry seed to seedlings with functional adventitious roots.

THE DRY SEED

Seed size (mass) has long been recognized as having an influence on seedling vigor. Generally, seed mass correlates well with seedling vigor within a species, but not between species (Wright 1971a). In comparing laboratory and field responses of two grasses, Asay and Johnson (1983) recently found that screening genotypes on the basis of seed weight still appears to be the best way to select for field performance. The use of seed size as a selection tool to increase seedling vigor within a species also has merit, as this procedure has yielded 'Verde', a kleingrass

(*Panicum coloratum* L.) cultivar with improved seedling vigor (Holt et al. 1983).

Unfortunately, potential seed mass of many genera is never achieved, chiefly because of seed abscission. Seed abscission in several warm-season grasses have been well described anatomically (Burson et al. 1978, 1983), but has been influenced little, if any, by the combined efforts of the physiologist and plant breeder. Plant growth regulators have not been completely effective in blocking this process (Weiser et al. 1979). Breeding for seed retention is difficult because many forage grasses are polyploids and seed retention can be a recessive character (McWilliam and Gibbon 1983). However, basic ecological information is needed to determine if seed shattering is advantageous or disadvantageous in an established pasture or range situation. We make the simplifying assumption that it would have no adverse affect on persistence and reseeding, but the critical experiments have never been performed. On the positive side, it is obvious that seed retention is a desired characteristic in seed production fields. Commercial producers will time their harvest to optimize seed yield; therefore, commercial seedlots always contain large percentages of light, immature seed. The data in Table 1 documents the percent composition by seed mass classes of a commercial seedlot of kleingrass purchased in Temple, Texas. Approximately 35% of the total mass of the seedlot was composed of individual seeds with a mass of 0.65 mg/seed or less:

Table 1. - Percent composition by seed mass classes of a commercial kleingrass seedlot

Fraction #	Seed mass/ 1000 seed (g)	Percent of total sample (by wt.)
1	0.285	5.62
2	0.369	7.76
3	0.652	19.69
4	0.740	27.99
5	0.840	9.30
6	0.896	23.64
7	1.010	6.00

We have experimentally demonstrated that seedlings from such seed are significantly less vigorous than seedlings from larger seed (Tischler, unpubl. results). The development of non-shattering genotypes would allow commercial producers to offer seed of more uniform maturity and seed mass, corresponding to fractions 4 and greater. Because the physiology and biochemistry of seed abscission in monocots is not well understood, the best approach at present may be to attempt to modify abscission genetically. The requisite genetic variation would have to be discovered through evaluation of new germplasm, interspecific hybridization with species having greater seed retention, or mutation breeding. Large populations and some form of inbreeding may be required to uncover recessive genes.

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THE IMBIBED, GERMINATING SEED

The previous discussion indicated the importance of seed size (and thus seed reserves) in influencing seedling vigor. In comparing seedling vigor of three grass species, however, we observed effects of mobilization of reserves on seedling vigor (Tischler and Voigt 1983a). Seed of kleingrass and wilman lovegrass (*Eragrostis superba* Peyr.) contain almost identical amounts of starch, but wilman lovegrass is much more vigorous at normal planting depths of 2-4 cm (Table 2). At deep planting depths, seedling vigor measured as shoot mass at 14 days post emergence of both grasses decreased, but the decrease was drastic in the case of wilman lovegrass:

Table 2. Relationship between planting depth and 14-day post emergence shoot weight in kleingrass and wilman lovegrass

Planting depth (cm)	Shoot mass	
	Kleingrass (n=311)	Wilman lovegrass (n=230)
2	0.138*	0.234
4	0.116	0.168
6	0.109	0.087
8	0.072	0.036

*Values generated from linear regression equations relating shoot mass to planting depth. R^2 values for both equations are significant at the 1% level of probability.

We measured rates of utilization of starch in seed of both species, and found that starch was metabolized much faster in wilman lovegrass seed. At deep planting depths, the wilman lovegrass seed were devoid of starch by the time the shoot emerged, while some reserves were still present in the kleingrass caryopsis. This observation suggested that screening for seedling vigor should be done at the depth at which the grass would normally be planted in the field situation because desirable genotypes may be discarded if screened by deep planting.

Most forage grasses produce seed which have some form of post-harvest seed dormancy. Also, in some cases, water-soluble germination inhibitors are present in the structures covering the caryopsis, and these compounds must be leached out before germination can occur. The usefulness of seed dormancy in grasses used for improved pastures has not been addressed. We have developed a non-dormant population of kleingrass, and will attempt to address that question in the near future. The almost universal presence of post-harvest seed dormancy in forage grasses as well as in many weedy grass species may be a survival mechanism that serves a necessary

function for the species at the plant community level. Interestingly, in kleingrass, dormancy is not an "all or nothing" phenomenon. Freshly harvested seed from individual plants will germinate from 1 to 20%, depending on the genotype involved (Tischler and Young 1983). Perhaps the plant is optimizing its chances for reproductive success by producing seed that is not completely dormant. We have no information on the relationship between dormancy level in a seedlot and the speed of germination of that portion of the seedlot which is not dormant. This is another question that needs further study.

The topic of speed or rate of germination has received considerable attention (Tucker and Wright 1965). However, the importance of speed of germination remains to be determined. When a field planting of a grass has been judged successful, we do not know if the early or the late germinators gave rise to the successful seedlings. We hypothesize that the early germinators were responsible, but the critical experiments have never been done. Very basic field ecological studies are needed to clarify such questions. Various treatments to speed up or synchronize germination have also received much attention. One such treatment involved soaking seed in water for 10-90 hours, and drying the seed before planting. This treatment produced a positive response in crested wheatgrass (*Agropyron desertorum* (Fisch. ex. Link) Schult.) (Bleak and Keller 1970). Numerous other papers described benefits of planting hydrated or pregerminated seed, but some problems were inherent in this technique (Hauser 1983). Other treatments involved the partial hydration and subsequent redrying of seed in an osmoticum such as polyethylene glycol (Bodsworth and Bewley 1981). This technique has shown promise with some vegetables (Heydecker and Gibbins 1978), but not with forage grasses (Tadmore et al. 1969). Osmotic agents have also been employed in attempts to screen for germination at low soil water potentials. This procedure is best done by enclosing seed and soil in a dialysis membrane, which is immersed in the osmoticum (Asay and Johnson 1983). After a specified interval, the germinating seedlings are removed and transferred to the greenhouse and eventually to the field for recurrent selection work. Controversy exists about the wisdom of planting seed which begin germinating when soil water is already in scarce supply. One can envision environmental situations where such behavior would be desired, but another set of conditions could result in a total failure of the planting. In this regard, Watt (1974) made the disturbing observation in comparing five grass species that the poorest establishers were the grasses which germinated at the lowest water potentials.

The above discussion suggests that seed in a seedlot may exhibit much phenotypic plasticity. Some of the seed germinate immediately, others are restricted by varying levels of dormancy, and still others germinate but do so only very slowly. Perhaps we need to consider the benefits of the maintenance of phenotypic plasticity in a

seedlot. Assuming that one had access to seedlots a) having no dormancy, b) having a high degree of dormancy, c) having been treated to accelerate germination, and d) having been selected for germination at low water potentials, perhaps the most fool-proof course of action might be mixing parts of each seedlot. The ratio used could be developed specifically for a given environment. This would serve as a kind of insurance against several sequential unfavorable weather scenarios. Evidence that this approach may have merit comes from data of Webster and Dahl (1983). They found that many weeping lovegrass seed that did not germinate initially following a field watering remained viable, and eventually gave rise to a vigorous stand after subsequent precipitation events.

THE EMERGED SEEDLING

Root Growth

Essentially all warm-season grasses exhibit the panicoid growth pattern, where adventitious roots arise from the crown of the plant, very near the soil surface. Until the shoot (and crown tissue) reach a sufficient size for adventitious root initiation, the primary root must supply water to the shoot. This fact is normally overlooked by crop physiologists, since only rarely do field crops such as sorghum experience drought in the seedling stage. In grasses, once the primary root had penetrated to a depth where soil water was readily available, the diameter of the metaxylem vessels in the subcoleoptile internode effectively limited the rate of water transport to the shoot (Wilson et al. 1976).

In our research on primary root development, we found that in kleingrass and weeping lovegrass, *Eragrostis curvula* (Schrud.) Nees, poor quality seed could give rise to seedlings in which the primary root was completely missing (Tischler and Voigt 1981). Thus, development of seed retaining genotypes would have a secondary benefit of reducing the number of these "no root" seedlings, which are doomed to failure in the field.

In studies where we documented the depth of penetration of primary roots, we found that primary roots of Catalina boer lovegrass, *E. curvula* var. *conferta* Nees selected for seedling drought tolerance (Wright 1971b), and Morpa lovegrass could penetrate (through a suitable soil media) to a depth of 50 cm or more.

Adventitious roots were clipped as they were initiated to restrict the seedlings to the primary root. The data presented in Table 3 demonstrate results from this experiment.

Table 3. Primary root length of Morpa and Catalina lovegrass at various times after planting

Days after planting	Primary root length (cm)		
	Morpa	Catalina	(Range)
7	2.0	2.4	1.1-3.5
14	5.9	4.4	2.0-14.6
21	16.3	14.5	7.2-28.3
42	40.3	40.0	23.2-56.1

The ratio of shoot mass to primary root length increased by at least a factor of three over the course of the experiment illustrating the increasing shoot size and corresponding transpiration load supported by a single primary root. The great range in values of primary root length for each sampling age was the principal problem with comparing differences in primary root length between genotypes. These grasses are apomictic; therefore, even more variability would be expected if one used this approach with sexual grasses. The data did demonstrate adequate capacity for primary root growth in these genotypes.

Shoot growth

When we compared mobilization of seed reserves in kleingrass and wilman lovegrass caryopses, we found that seed reserves were depleted by 6 days after planting (Tischler and Voigt 1983b), which agreed well with data for blue panicgrass (*Panicum antidotale* Retz) (Wright 1980). In contrast, seed mass (seed reserves) continued to decline in wheat until 11 days after planting (Williams 1960). This indicates that forage grass seedlings must become self sufficient at an earlier stage of development than larger seeded field crops. We found that 6 days after planting kleingrass and wilman lovegrass seedlings were capable of reducing nitrate to ammonia, and contain appreciable levels of chlorophyll (0.84 and 0.73 mg/g fresh weight, respectively (Tischler and Voigt 1983b). Although the capacity for autotrophic growth develops rapidly in warm-season forages, screening and selection for greater physiological or biochemical efficiency should ultimately lead to genotypes with faster growth rates. Evidence that such possibilities exist comes from two sources. The first was a report by Clements and Latter (1973) that selection for heavy seedlings in *Phalaris tuberosa* did not significantly increase seed weight. A second bit of evidence comes from preliminary experiments in which the analog of a plant steroid was used. This compound, called brassinosteroid, has been shown to speed growth rate in "slow" barley seedlings (Gregory 1981). We treated kleingrass and wilman lovegrass seed with this compound (100 ppm in methylene chloride), and observed significant increases in 15-day post-emergence shoot mass in kleingrass, but not in wilman lovegrass (Tischler and Voigt, unpubl. observations). We do not know if the compound speeds the rate of mobilization of seed

reserves, or rather modifies metabolism of the shoot. Results of this study have encouraged us to try other modes and rates of application of brassinosteroid and to test the material on other species. By speeding shoot growth, this compound could artificially improve seedling vigor.

INITIATION AND GROWTH OF ADVENTITIOUS ROOTS

Briske and Wilson (1978) have outlined the requirements for adventitious root initiation and elongation in blue grama. Briefly, these factors include a favorable water balance in the shoot and favorable temperatures. We suspected that a shoot size requirement was also involved in the case of kleingrass and wilman lovegrass. If a minimum shoot mass is required for adventitious root initiation and growth, then screening for shoot mass would be a direct and easy way to improve establishment. However, if adventitious root initiation is somewhat independent of shoot mass, the selection could be made only by excavating individual plants and measuring root lengths. We addressed this question, and found that, in general, age of the seedling was more important than shoot mass in determining when adventitious roots were initiated (Tischler et al. 1983). Our experience has been that characterization of adventitious root parameters was difficult because of the tremendous plant-to-plant variability in these characteristics. Not only was variability observed in age or seedling mass at adventitious root initiation, but also in number of roots initiated. Apparently, there are no direct methods to circumvent these experimental difficulties.

In summary, many interesting avenues of research could ultimately lead to the improvement of establishment in warm-season forage grasses. Unfortunately, many of the most elementary or logical types of studies have not been done. A more thorough understanding of the interaction of the seed and seedling with the physical environment would allow plant scientists to devise more appropriate screening systems for improving seedling establishment. While some selection procedures may appear beneficial in controlled environment studies, they must ultimately produce beneficial results in the field. Plant characteristics that are easy to quantitate and could theoretically improve establishment may not necessarily be beneficial to the plant in the environment where it must ultimately perform.

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INTRODUCTION

Production and quality of pure grass pastures can be improved by seeding into the existing sod a legume such as ladino clover (Trifolium repens L.).

Competition from the existing sward may be an important factor limiting establishment and growth of sod-seeded forage legumes (Wilkinson and Gross 1964). Herbicidal suppression of grass improves establishment of sod-seeded legumes under some conditions. Fribourg et al. (1978) enhanced stands and yields of clovers sod-seeded into tall fescue (Festuca arundinacea Schreb.) by chemically suppressing the grass during establishment. Banding paraquat (1,1'-dimethyl-4,4'-bipyridinium ion) over the row to be seeded increased stands and size of alfalfa (Medicago sativa L.) and ladino clover seedlings when sown into dense and vigorously growing Kentucky bluegrass (Poa pratensis L.) (Taylor et al. 1969). In another study, paraquat banded over the seeded row had little effect on stand density but was found to consistently enhance development of alfalfa and clover seedlings in a Kentucky bluegrass sod (Taylor et al. 1972).

Damage by insect pests on emerging stands of legumes seeded into grass swards has been observed, however, little detailed information is available. Other workers (Fribourg and Stanley, 1960) have indicated that insects may play an important role in the persistence of established stands of ladino clover. Hoveland et al. (1966) in Alabama showed that stand and yield losses of sod-seeded annual clovers was probably caused by pygmy crickets [Nemobius fasciatus (De Geer)]. Snails [Polygyra cereolus (Muhlfeld)] (Kalmbacher et al. 1979) and slugs [Agriolimax reticulatum (Muller)] (Welty et al. 1981) also limit establishment of sod-seeded forage legumes.

Limited data are available on insect problems associated with fall sod-seeding of legumes with no reports of successful mid-to late-fall plantings in sods. The purpose of these studies was to determine the effects of seeding date, grass suppression, and insects on fall and spring establishment of ladino clover and spring establishment of alfalfa into tall fescue sods. (No insect controls in spring studies.)

MATERIALS AND METHODS

For the fall studies, four field experiments (exp.) of 1-year duration each were conducted from 1978 to 1981 on separate pasture sites predominated by tall fescue (75 to 95% fescue cover) in Piedmont North Carolina.

For the spring studies, seven separate field experiments were conducted in the Piedmont of North Carolina from 1978-81. Although locations were different each year, alfalfa and clover experiments were adjacent except in 1979 when only clover was planted.

Studies were conducted on a Cecil (clayey, kaolinitic, thermic Typic Hapludult) both clay loam and sandy loam and on an Enon (fine, mixed, thermic Ultic Hapludalf) fine sandy loam. Soil reaction for all sites about 16 months prior to seedling ranged from pH 6.1 to 6.4 and the P and K levels were above medium. Dolomitic limestone, P, and K were added to all sites 2 to 6 weeks prior to seeding according to soil test recommendations. The fescue sod was clipped to a 4-cm stubble height and the clippings were removed at 7 days (d) and again at 1 d prior to fall seedings and in October and again 1 d prior to seeding for the spring seedings.

Seed and Chemical Application

'Tillman' ladino clover seed was inoculated with Rhizobium trifolii using a water-syrup sticking agent and drilled into rows spaced 25.4 cm apart at a rate of 5.2 kg ha⁻¹ using a Tye Pasture Pleaser sod seeder for the fall seedings. For the spring seedings, ladino clover was sown at 5.6 and alfalfa at 22.4 kg ha⁻¹. Plots were 2.0 m (8 rows) wide by 7.1 m in length and treatments were arranged in a randomized complete block design with either four or five replications. Smaller sized plots were used for the spring experiments. A power dethatcher with tines spaced 20 cm was used to produce a slot in the sod about 2 cm in depth and plots were seeded with a "Planet Jr." push-type seeder. Paraquat for sod suppression and a granular carbofuran (2,3-dihydro-2,2-dimethyl-7-benzofuranyl methylcarbamate) for insect control was not applied or were applied at 0.28 and 3.7 kg ha⁻¹ a.i., respectively, at two fall seeding dates [early-September (Date 1) or mid-October (Date 2)] in a 2³ factorial combination. Only paraquat (no insecticide) was used in the spring. The two spring planting dates were mid-February and mid-March. Paraquat was broadcast in either 187 or 234 L ha⁻¹ of H₂O just prior to seeding using a CO₂ pressurized backpack sprayer. Carbofuran granules were applied directly in the drill row at 1.5 kg ha⁻¹ a.i. using a supplemental small seed hopper on the sod-seeder and by broadcasting the remaining 2.2 kg ha⁻¹ a.i. at seeding.

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Stand Determinations

Clover stands were determined soon after emergence by counting the plants in six (spring studies) or eight (fall studies) random 30-cm segments of row per plot. Also, a line transect method was used to estimate gross changes in clover population from fall to spring. In fall studies, an assessment of winter damage (stand survival), particularly for the Date 2 seeding, was made by counting numbers of plants located in two fixed quadrants 0.5 m in length in each plot.

Stage of growth of clover plants was determined between 1 and 5 December in each fall experiment by measuring the number of trifoliolate leaves present on each of 100 plants sampled per plot in Date 2 check plots.

Dry Matter Yields

Dry matter yields were determined by making periodic harvests. Visual estimates of percent clover, grass, and weeds were made prior to each harvest. The forage was dried at 57°C for 48 hr and weighed.

RESULTS AND DISCUSSION

Fall Establishment Studies

Growth stages of morphological development measured in December for the fall mid-October (Date 2) ladino are shown in Table 1 with minimum air temperatures. Good winter survival was obtained from October sod-seedings although the majority of clover seedlings (75 to 83%) had developed only one trifoliolate leaf by early December in three of four trials.

Ex.	Check					Stage of growth ¹ P					Minimum winter air temperatures	
	Trifoliolate	1	2	3	4+	Trifoliolate	1	2	3	4+	Range	Days
1	13	67	16	4	0	--	--	--	--	--	-10 to	-13 8
2	12	73	13	2	-	--	--	--	--	--	-10 to	-14 11
3	2	22	60	15	1	1	10	45	40	4	-10 to	-11 5
4	21	73	6	0	0	10	64	26	0	0	-10 to	-16 9

¹P = paraquat (broadcast), U = unifoliate stage; stage of growth measurement made 1-5 December.

Insect control and sod suppression were found to enhance and, in some cases, be essential to establishment of ladino clover into a tall fescue sod. Insects can cause extensive clover stand losses within 5 d of seedling emergence, thus demonstrating the importance of making early and detailed observations to determine the causes of stand losses. During a 9-day period shortly after the September seeding in one experiment an 82% reduction in clover density was measured in plots not

treated with insecticide. The degree of insect damage varied considerably among experiments. However, in general less insect damage occurred in Date 2 seedlings because of the reduced numbers of insects in October compared to September.

In contrast to the findings of this study, Mangan *et al.* (1982) noted only slight effects of pesticide treatments on legume seedling survival during the first month of establishment (white clover was not included).

Suppression of the fescue sod using paraquat also increased stands and yields of ladino clover, probably by reducing competition for light and moisture. Application of paraquat at 0.28 kg ha⁻¹ in October in this study generally gave excessive sod suppression (90% permanent kill) with poor fescue recovery. The tendency for excessive suppression of fescue (for ladino pastures) in October would likely be alleviated by band application. Measurements in December of fescue recovery growth from September application of paraquat showed a range of 35 to 70% permanent damage to the fescue.

Increases in clover stands and yields also were obtained by the combined application of P and I. Under conditions of limited moisture availability and high insect populations, the use of P and I together appears necessary to produce sufficient stands and growth of ladino clover to permit maximum expression of the effects of each treatment. In one experiment, seasonal yields of September-seeded clover were 2,565, and 1,080 kg ha⁻¹ for paraquat and carbofuran used alone, respectively, and 5,570 kg ha⁻¹ when used in combination. Check plots yielded 185 kg ha⁻¹. By early October in all experiments the fescue regrowth in Date 1 plots receiving I but not treated with P had reached heights ranging from 10 to 15 cm. Etiolated and dying seedlings of ladino clover were observed in those plots due probably to competition for light and moisture by the sod.

Spring Establishment Studies

Drilling legume seeds vs surface placement resulted in two to four times as many seedlings initially established and up to 2,000 kg ha⁻¹ more legume yield during the first season. Nonetheless, satisfactory stands were eventually obtained from surface seeding ladino in February in 3 of 4 years. Planting in February resulted in better stands and yields of clover than March in 3 of 4 years.

Using paraquat for sod-suppression often resulted in significant advantage when clover was seeded at the later date (March). Alfalfa produced very low yields during the first season under all treatments (usually one or two harvests) and did not appear to compete well with the tall fescue sod when spring seeded. The fescue growth did not appear to

be affected by the February application of paraquat and only temporary grass suppression was noted from March applications. The highest first year alfalfa yields obtained during these studies ranged from 795 to 4,610 kg ha

SUMMARY

We conclude that the use of both an insecticide and grass suppressant may be necessary for successful establishment of ladino clover into a tall fescue sward under some conditions (particularly in September seedings) and that mid-October is a good alternative to early-September as a seeding date during years of low moisture and high insect populations in September.

February and March are satisfactory alternative dates for seeding ladino clover into a fescue sward, but not for alfalfa with present techniques.

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The use of trade names in this publication does not imply endorsement by the North Carolina Agricultural Research Service of the products named nor criticism of similar ones not mentioned.

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Apomixis has been reported in approximately 150 different grass species and is prevalent in many of the tropical and subtropical species. Because apomixis is a vegetative means of reproduction in which an embryo is formed without the union of an egg cell with a sperm nucleus, there is no genetic variability expressed in the resultant progeny. They are identical to the apomictic parent. Apomixis has been a major impediment to the improvement of some apomictic species and a valuable tool in others.

The approaches to improving apomicts have varied, depending upon the species. In obligate apomicts without known sources of sexuality, the only method of improvement is to select among the variable apomictic ecotypes that occur in most apomictic species. This has been the most common approach used by plant breeders.

When a source of sexuality is discovered in an otherwise obligate apomictic species, it provides an opportunity to improve the species through hybridization with the apomictic ecotypes. Highly vigorous, heterozygous apomictic F_1 hybrids are often produced. An added advantage of obligate apomictic hybrids is permanently fixed heterosis which will be expressed indefinitely in future generations. This approach has been successful in the development of improved buffelgrass, *Cenchrus ciliaris* L., cultivars (Bashaw 1968, 1980).

Many species are facultative apomicts, i.e., their individual plants can reproduce by both sexual and apomictic means. The amount of sexual or apomictic reproduction is apparently dependent upon the genotype of the plant. Because of the more frequent occurrence of sexuality in a facultative apomicts, new genotypes are continually produced. This source of variation can be used for improvement of the species. With the realization that facultative apomixis is more common in many of the warm-season grasses than was once thought (Voigt and Bashaw 1976, Sherwood et al. 1980), philosophies concerning the breeding of apomictic grasses have been altered during the past few years.

Apomixis is prevalent within the genera *Cenchrus*, *Eragrostis*, and *Paspalum*. Because of

new findings regarding apomixis in members of these three genera, different approaches have been developed to improve them. The purpose of this paper is to summarize some of the different approaches being used to improve buffelgrass, lovegrass, and dallisgrass.

BUFFELGRASS

Buffelgrass was once considered an obligate apomict. Improvement was limited to selecting among existing apomictic ecotypes until a sexual mutant was discovered in a seed production field in 1958 (Bashaw 1962). Using this sexual plant, Taliaferro and Bashaw (1966) determined the inheritance and genetic control of apomixis in buffelgrass. They also proposed a successful hybridization scheme in which the sexual plant is crossed with apomictic plants to produce apomictic true breeding F_1 hybrids that are new potential cultivars (Figure 1).

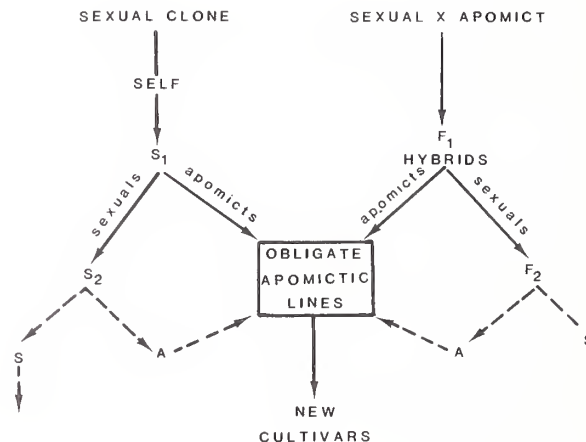


Figure 1. Diagram showing the development of new cultivars using sexual and obligate apomictic buffelgrass.

As additional germplasm was obtained and evaluated, facultative apomixis was found in the species (Sherwood et al. 1980). In 1976 a plant exploration trip to South Africa was conducted to collect new buffelgrass germplasm. More than 800 ecotypes were collected of which 733 were obligate apomicts and 73 were facultative apomicts. No obligate sexual plants were collected, but about 5% of the sexual offspring from selected facultative apomicts are obligate sexuals. This has provided new sources of sexual germplasm for the buffelgrass hybridization program. However, many of the sexual plants are weak, sterile aneuploids and considerable research is needed to determine the best sources of sexual material.

A major purpose of the collection trip was to secure germplasm with improved winter hardiness. A group of 49 nonrhizomatous accessions, all with 45 chromosomes have more winter hardiness than

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any other buffelgrass germplasm. Buffelgrass normally has 36 chromosomes. During meiosis the extra 9 chromosomes are present as 9 univalents. These extra chromosomes undoubtedly represent an alien genome from another species. The 45-chromosome plants probably originated from the fertilization of an unreduced buffelgrass egg by pollen from an unknown diploid species.

Six of the 45-chromosome plants are facultative apomicts and the remaining 43 are obligate apomicts. Cytological studies of the ovules of the facultative apomicts, showed that the percentage sexual embryo sacs ranged from 5 to 50; however, only 1 to 3% of the selfed progeny were off-types. Apparently a high level of abortion occurs in the sexual sacs. Pollen quality in these accessions is also low and hybridization is difficult. However, 21 hybrids were recovered from crosses between a sexual 36-chromosome genotype and two of the more vigorous 45-chromosome accessions. Chromosome number of the hybrids ranged from 36 to 45. The 45-chromosome plant is an obligate apomict and the others are sexual. The only hybrid that survived the severe 1983-84 winter was the 45-chromosome plant. This suggests that the genes for cold tolerance are on the 9 extra chromosomes.

Because of the sterility problems in these new accessions, some of the more traditional approaches to hybridizing apomictic buffelgrass have been modified. The more sexual and fertile facultative apomictic 45-chromosome accessions are being used as female parents and crossed with the highly rhizomatous apomictic cultivar 'Llano' in an attempt to increase cold tolerance by combining tissue tolerance with rhizomes. This approach will provide the possibility for hybridization in two ways: (1) normal fertilization of an egg in a sexual embryo sac and (2) fertilization of an unreduced egg in an apomictic sac.

A technique that has increased the effectiveness of the 45-chromosome accessions as pollen sources is collection of the inflorescences early in the morning prior to anthesis and keeping the culms in water in a refrigerator until pollination time. The cold temperature in the refrigerator maintains a higher pollen viability than when the inflorescences are kept in water in the greenhouse.

LOVEGRASS

Weeping lovegrass, *Eragrostis curvula* (Schr.) Nees is a polymorphic species that reproduces by diplosporous apomixis (Streetman 1963). It was once considered an obligate apomict, but sexual plants were discovered (Voigt and Bashaw 1972) and later it was determined that some plants are facultative apomicts (Voigt and Bashaw 1976). The *Eragrostis curvula* complex is composed of

several diverse types which include weeping and boer lovegrasses as well as the larger robusta types of weeping lovegrass.

Sexual or highly sexual plants are used as female parents in crosses with apomictic plants. A cross between a sexual weeping lovegrass type ($2n=40$) produced progeny which were classified as 40% sexual, 20% intermediate, and 40% apomictic based on cytological observation of the embryo sac development. The same hybrids were classified in the field by progeny test as 50% sexual, 10% intermediate, and 40% apomictic (Voigt and Burson 1983). This demonstrates that sufficient numbers of new apomictic hybrids are produced from such crosses to allow progress in plant improvement by using an apomictic breeding scheme as illustrated in Figure 2.

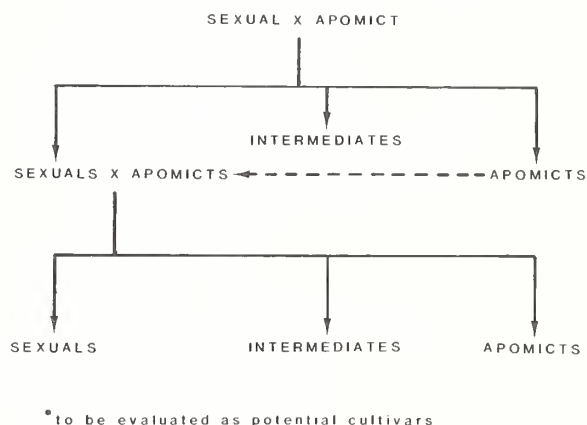


Figure 2. Breeding scheme used for improved facultative apomictic species.

When several different sexual tetraploids were crossed with the robusta type Renner ($2n=60$), the progenies were classified as 53% sexual, 9% intermediate, and 38% apomictic. In crosses using the same sexual females, and another robusta type, R-1 ($2n=69$), the hybrids were classified as 9% sexual, 4% intermediate, and 87% apomictic, a ratio of 9.6 apomicts to 1.0 sexual. Of the apomictic hybrids recovered, 75% were considered sufficiently uniform for potential use as apomictic cultivars (Voigt and Burson 1983).

The most important agronomic objective for developing an apomictic breeding scheme to be used in improving weeping lovegrass is to improve forage quality. Variation for digestibility within existing winter hardy plant collections is low (Voigt et al. 1978). Variation within the less hardy robusta type germplasm is greater (Voigt and Tischler 1984). While apomictic breeding using boer X weeping lovegrass hybrids shows some potential for improving digestibility (Voigt 1984), hybridization with more digestible robusta

germplasm should have even more potential. It is obvious that apomixis is no longer a barrier to the improvement of weeping lovegrass, but is now an important tool for the improvement of the E. curvula complex.

DALLISGRASS

Common dallisgrass, Paspalum dilatatum Poir, is a obligate apomict that has 50 chromosomes (Bashaw and Holt 1958) and cytologically is somewhat similar to that of the 45-chromosome buffelgrass accession. The 50 chromosomes pair as 20 bivalents and 10 univalents at meiosis. Unlike many obligate apomicts, there are very few apomictic biotypes with chromosome numbers of $2n=40$ and 60 and a sexual yellow-anthered biotype, $2n=40$ (Bashaw and Forbes 1958; Bashaw and Holt 1958; Moraes Fernandes et al. 1968). Unfortunately, the sexual biotype has contributed little to the direct improvement of common dallisgrass because of the difficulty in crossing the two biotypes and it was a poor forage type. The hybrid has 45 chromosomes which pair as 20 bivalents and 5 univalents (Bennett et al. 1969). This plant has provided valuable basic information and provides a possible means for the manipulation of apomixis in common dallisgrass. Several different approaches are currently underway to manipulate apomixis and facilitate improvement of the species.

Chromosome pairing in the F_1 dallisgrass hybrid revealed that the 40 chromosomes of the sexual yellow-anthered biotype are homologous to 40 of the 50 chromosomes in common dallisgrass. This information provided a basis for phylogenetic investigations using the sexual biotype in various crosses to obtain information that could be extrapolated to common dallisgrass (Burson 1983). The rationale for this program is to identify the natural progenitors of common dallisgrass and synthesize sexual types by crossing the progenitors. Two of the genomes in common dallisgrass have been identified (Burson 1983).

The lack of apomictic reproduction and the poor forage characteristics of the yellow-anthered biotype and the F_1 hybrid suggests that a number of valuable genes occur on several of the extra chromosomes in common dallisgrass. Therefore, if the 10 univalents were stabilized in a sexual plant, improvement of common dallisgrass should be possible. In an effort to accomplish this, the sexual F_1 hybrid has been backcrossed with common dallisgrass. Eighteen BC_1 plants have been recovered. They will continue to be backcrossed with common dallisgrass until all of the univalents have their homologs transferred into the BC plants. This should result in plants with 58 to 60 chromosomes depending upon when apomixis is expressed in the BC plants. If a promising apomictic type is recovered, it will be selected and evaluated as a potential cultivar.

Another similar approach involves crossing the sexual F_1 hybrid with the new apomictic 60-chromosome accessions from South America (Burson 1984). This cross could provide a more efficient method for stabilizing the univalents because of the meiotic stability in the 60-chromosome plants. Hybrids from this cross should also provide valuable information concerning the cytological composition of the 60-chromosome biotype. Crosses have been attempted between the sexual yellow-anthered biotype ($2n=40$) and the apomictic 60-chromosome biotype. This could provide new information regarding apomixis as well as the origin of common dallisgrass.

A more unconventional approach would be to use apomictic common dallisgrass as the female parent in crosses with the apomictic 60-chromosome biotype. Hybrids would be derived by the fertilization of an unreduced egg. Even though the frequency of such crosses would be low, the possible gains from a hybrid between these two biotypes could be worth the effort, and some interesting genotypes could result.

Sexual diploids have been discovered in plant introductions of P. plicatulum Michx. and P. alcalinum Mez. recently collected in South America. All other biotypes of both species are tetraploid obligate apomicts. Efforts are underway to double the chromosome number of both of these diploids. If we are successful in doubling their chromosome numbers and maintaining sexuality, it will be possible to improve both grasses by crossing the sexual plants with the apomictics. This will be similar to the original approach used in improving buffelgrass.

SUMMARY

During the past 15 to 20 years, philosophies regarding improvement of apomicts and the approaches used in the breeding of apomictic forage grass species have changed. This has been brought about primarily because facultative apomixis is more prevalent in the warm-season grasses than was once thought. The use of highly sexual facultative plants rather than obligate sexual plants the female parent in crosses with apomictic types does not yield as conclusive genetic data on mode of reproduction in the progeny, but this approach is effective in producing improved apomictic forage types in lovegrass and buffelgrass. Current methods being used to manipulate the chromosomes controlling apomixis require detailed cytogenetic analysis and these are discussed. Because of the similarities in the cytology of common dallisgrass and the 45-chromosome buffelgrass accessions, the approaches used to manipulate apomixis in one of these species will apply to the other.

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INTRODUCTION

Plant regeneration from callus cultures has been reported for all major gramineous species. However, until recently the development of plant tissue culture technology for grasses and cereals has lagged far behind that of many other agronomic crops because plant regeneration has frequently been of low frequency and of short duration (see Conger 1981 for review). These problems are being increasingly circumvented by attention to culture media components, use of certain responsive cultivars and genotypes, and careful visual inspection of cultures to select for promising material (Nabors et al. 1983). Also, recent work (Vasil and Vasil 1982a) suggests that the phenomenon of nonzygotic (somatic or adventive) embryogenesis may be commonplace in the Gramineae, providing great optimism that emerging *in vitro* technologies can be employed for crop improvement. This presentation will focus on nonzygotic embryogenesis in forage grasses including those grain producing species such as *Zea mays* L. and the millets that are also commonly used as forage. Results from our ongoing experiments with *Dactylis glomerata* L. will be employed for illustrative purposes.

HISTORY

Nonzygotic embryogenesis was first documented in carrot by Stewart (1958), Stewart, Mapes and Mears (1958) and Stewart, Mapes and Smith (1958) who suggested that certain cell cultures grew in a manner "strangely reminiscent of the early stages of embryological development from the egg and proembryo." Since Stewart's pioneering work, nonzygotic embryogenesis has been documented in cultures of many other species (see Tisserat et al. 1978 for review). The first suggestion that this phenomenon also occurred in the Gramineae was when Gamborg et al. (1970) dispersed callus-derived cell clumps of *Bromus inermis* Leyss into liquid medium to obtain albino plantlets via somatic embryogenesis. The next report in grasses was when Thomas et al. (1977) described embryo-like structures arising from the scutellum of immature zygotic embryos of *Sorghum bicolor* (L.) Moench with many green plants recovered. A clearer description of the phenomenon was provided by Vasil and Vasil

(1982b) who correlated light microscopy with scanning electron microscopy to study nonzygotic embryo ontogeny in *Pennisetum americanum* (L.) K. Schum. Nonzygotic embryogenesis has now been reported for several agronomically important forage grasses (selected literature summarized in table 1). In addition, it is likely that some earlier reports of regeneration from grass tissue cultures may have also occurred by a then unrecognized process of embryogenesis.

CONSIDERATIONS FOR OBTAINING EMBRYOGENIC CULTURES

Several factors are potentially important for inducing embryogenesis in grass tissue cultures including genotype, medium and explant sources. There is conflicting evidence as to whether or not nonzygotic embryogenesis is under genetic control. For instance, in *Z. mays* the response originally occurred in only a few genotypes (Green et al. 1983). Workers (Lu et al. 1982) have since documented this phenomenon in additional lines. However, these were hybrid cultivars of unknown pedigrees. Therefore, it is possible that each of these cultivars were highly related. Our observations with *D. glomerata* (unpublished) demonstrated that only seven percent of 330 genotypes tested showed an embryogenic response.

A number of media have been successfully employed to obtain embryogenic cultures. These include modifications of MS (Murashige and Skoog 1962), SH (Schenk and Hildebrandt 1972) and N-6 (Chu et al. 1975). Various additives such as casein hydrolysate, inositol, L-proline and other amino acids were sometimes included. Sucrose has been commonly added at two to three percent although some reports (Green et al. 1983) indicate a stimulation of embryogenesis at elevated levels of six to nine percent. Several auxins have been used to produce an embryogenic response with 2,4-D being the most common. Dicamba (Hanning and Conger 1982) and 2,4,5-T (Heyser and Nabors 1982) are also effective. Weaker auxins such as IAA and NAA are relatively ineffective. Cytokinins may actually inhibit embryogenic responses in some species (personal observation).

Immature zygotic embryos remain the most commonly used explants for obtaining embryogenic culture (Table 1). However, leafbase meristem and young inflorescence explants have been successful with several species. There has been a trend to extend the embryogenic response to other explants including anthers, mature embryos, mesocotyls, nodes, pistils, seeds and stems (Table 1). Although nonzygotic embryogenesis in *Z. mays* is still achieved only by use of immature embryos, promising experimentation (Chang 1983) indicates that the leaf-bases of *in vitro*-germinated immature embryos may also give rise to embryogenic cultures.

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Table 1. Chronology of selected nonzygotic embryogenesis literature for forage grasses

Date	Species	Reference(s)	Explant (Culture-type)
1970	<u>Bromus inermis</u>	Gamborg et al.	mesocotyl (callus)
1973	<u>B. inermis</u>	Kao et al.	suspension (protoplast)
1977	<u>Sorghum bicolor</u>	Thomas et al.	immature embryo (callus)
1980	<u>Lolium multiflorum</u>	Dale	immature embryo (callus)
	<u>Pennisetum americanum</u>	Vasil and Vasil	immature embryo (callus to suspension to protoplast)
	<u>S. bicolor</u>	Wernicke and Brettell	leaf (callus)
1981	<u>L. multiflorum</u>	Dale et al.	inflorescence, node (callus)
	<u>Panicum maximum</u>	Lu et al. Lu and Vasil (a) Lu and Vasil (b)	suspension (protoplast) immature embryo, inflorescence (suspension) leaf (callus)
	<u>P. americanum</u>	Vasil and Vasil	immature embryo (callus to suspension)
	<u>Pennisetum purpureum</u>	Haydu and Vasil	anther, inflorescence, leaf (callus)
	<u>P. americanum</u> x <u>P. purpureum</u> hybrid	Vasil and Vasil (b)	immature embryo, inflorescence (callus)
1982	<u>Dactylis glomerata</u>	Hanning and Conger McDaniel et al.	leaf (callus, direct embryogenesis) mature embryo (callus)
	<u>Panicum miliaceum</u>	Heyser and Nabors	immature embryo, leaf, mesocotyl, seed, stem (callus)
	<u>P. americanum</u>	Vasil and Vasil	inflorescence (callus to suspension)
	<u>P. purpureum</u>	Wang and Vasil	inflorescence (callus)
	<u>Zea mays</u>	Genovesi and Collins Lu et al.	anther (direct embryogenesis) immature embryo (callus)
1983	<u>D. glomerata</u>	Conger et al.	leaf (callus, direct embryogenesis)
	<u>Panicum miliareci</u>	Rangan and Vasil	inflorescence (callus)
	<u>P. purpureum</u>	Vasil and Vasil	inflorescence (callus to suspension to protoplast)
	<u>Schizachyrium scoparium</u>	Songstad	inflorescence (callus)
	<u>Z. mays</u>	Green et al.	immature embryo (callus to suspension)
1984	<u>D. glomerata</u>	Gray et al.	nonzygotic embryo (callus, suspension)
	<u>D. glomerata</u>	unpublished	anther, pistil (callus, direct embryogenesis)

TYPES OF EMBRYOGENIC CULTURES

Table 1 summarizes in parentheses the type of embryogenic cultures obtained from explants of given forage species. Embryogenic callus cultures are most common and are generally characterized as being white to off-white and slow growing. The cultures are heterogeneous in composition with friable and nonfriable regions. An abundance of roots and root primordia are frequently present. Embryos usually arise from nonfriable cell masses. In addition, a mucilaginous material, probably of pectinaceous composition (Conrad et al. 1982), is frequently associated with the cultures. Cultures of D. glomerata are highly friable and embryos form both on and embedded in the callus mass. Embryos and/or embryogenic calli can be removed and placed on medium without auxin to induce germination. Embryos that remain in the presence of auxin eventually become disorganized to form more embryogenic callus. Thus, there is a cyclic process of organization to disorganization of embryos (Gray et al. 1984).

In contrast to callus cultures, the establishment of embryogenic cell suspension and protoplast cultures has been much more difficult in the Gramineae. Embryogenic cell suspension cultures have been reported for only four forage grasses (Gray et al. 1984, Green et al. 1983, Lu and Vasil 1981a, Vasil and Vasil 1981a). An early report (Gamborg et al. 1970) obtained embryos in liquid medium but not from true suspension cultures that could be routinely transferred and propagated. Embryogenic suspension cultures all tend to grow as clumps although friability of Z. mays suspension was enhanced by the addition of 0.1 μ M abscisic acid (Green et al. 1983). Proembryoids formed in these cultures and, except for D. glomerata, had to be physically removed and plated onto various solidified media for further development. Nonzygotic embryos of D. glomerata developed to a stage capable of germination directly in suspension cultures (Gray et al. 1984). Germination was induced either sterilely in liquid or on solid medium lacking auxin or under nonsterile conditions in water or sand.

Although nonzygotic embryogenesis has been reported from protoplasts of four gramineous species (Kao et al. 1973, Lu et al. 1981, Vasil and Vasil 1980, Vasil et al. 1983), the establishment of mature plants has yet to be documented for any grass species. This represents the greatest remaining roadblock towards applying in vitro technologies in the Gramineae.

Nonzygotic embryogenesis from tissue cultures of haploid floral parts is potentially useful for developing pure breeding lines (Wu et al. 1983), especially in highly heterozygous outcrossing species (eg. Schizachyrium scoparium (Michx.) Nash and D. glomerata). Plants possessing gametic chromosome numbers have been obtained from inflorescence cultures

of Festuca arundinacea Schreb. (Kasperbauer et al. 1980) but not via embryogenesis. However, nonzygotic embryogenesis from dividing microspores of Z. mays resulted in haploid plants (Genovesi and Collins 1982). Our unpublished experiments with D. glomerata showed direct embryogenesis from the outer anther wall and from within anthers. In addition, embryos arose directly from the ovary, style and stigma of cultured pistils. However, none of our regenerated plants have yet been shown to possess the gametic chromosome number.

NONZYGOTIC EMBRYO ONTOGENY

The study of nonzygotic embryo ontogeny can facilitate our understanding of embryogenesis in general. Because nonzygotic embryogenesis occurs within the obscuring tissues present in seeds (eg. endosperm and integuments), the process can be monitored more closely. A zygotic or nonzygotic plant embryo can be defined as a new individual arising from a single cell and having no vascular connection with maternal tissues (Haccius 1978). However, nonzygotic embryos often appear to deny the rule of unicellular origin because they seem to arise by a process of budding from pre-existing tissue. But, the pre-existing tissue is actually a proembryonal complex which was itself formed from a single dividing cell and this process is termed cleavage polyembryony (Haccius 1978). A similar type of embryogenesis occurs naturally in some species of gymnosperms (Buchholz 1933) and primitive angiosperms (Haccius and Bhandari 1975). In fact, the specialized suspensor of zygotic embryos can be regarded as "evolved" and rudimentary proembryonal complex. Many species which normally form suspensors during zygotic embryogenesis tend to develop enlarged proembryonal complexes in cell cultures. Thus, a trend to a more primitive evolutionary state of embryogeny seems to often be induced in tissue culture.

The process of nonzygotic embryogenesis has been described in immature zygotic embryo explants of P. americanum (Vasil and Vasil 1982b) and can be summarized as follows. After plating onto suitable medium, cells near the main procambial strand in the scutellar node region became densely cytoplasmic and separated from each other. Divisions in these separated cells eventually lead to the formation of nonzygotic embryos, basally attached to proliferating scutellar cells. The presence of suspensors was not documented but the single cell origin of the embryos was confirmed.

Histological studies of embryogenic leaf-base meristems of D. glomerata have not yet demonstrated a similar single cell origin directly (Conger et al. 1983), however proembryonal complexes that develop from mesophyll cells and from which many nonzygotic embryos arise suggests such an occurrence. Embryos of D. glomerata either developed from a callus or arose directly from leaf mesophyll

regions (Conger et al. 1983).

Embryo ontogeny in the direct response is currently under investigation in our laboratory. The developmental morphology of the nonzygotic embryos is similar to what Brown (1960, 1965) described for zygotic grass embryos. Results of our ongoing research can be summarized as follows. During early development, an embryo ruptures the leaf epidermis and assumes a globular shape on the surface. Cells of the embryo are all relatively densely cytoplasmic at this stage and are undifferentiated from each other. Next, the embryo elongates, becomes clavate and a degree of cellular differentiation is present. At this stage, large vacuolate, starch-filled cells which are destined to compose the scutellum, as well as a region of densely cytoplasmic cells which are relatively surface oriented and will develop into nonscutellar tissues are evident. The embryos elongate further and organized divisions of densely cytoplasmic surface cells create a notch which marks the future position of the coleoptile and shoot apex. As the scutellum enlarges by divisions of surface cells, the coleoptile develops outwardly by similar surface divisions and eventually closes the notch, creating a cavity within which the shoot apex differentiates. The radicle develops endogenously in tissue basal to the shoot apex eventually resulting in a complete embryonal axis. Scutellar cells of fully developed nonzygotic embryos contain an abundance of starch and lipids. Coleoptile cells appear to contain only starch whereas cells of the embryo axis possess only lipids. Dactylis glomerata embryos that arise directly from leaves often possess a well defined suspensor whereas those that grow from callus cultures usually have a broader connection to underlying tissue as is seen with nonzygotic embryos of other grass species.

USES OF NONZYGOTIC EMBRYOGENESIS FOR CROP IMPROVEMENT

Nonzygotic embryogenesis offers several advantages when compared to other methods of plant regeneration. Not only are higher yields of regenerants obtained but problems of long-term culture totipotency that are often encountered in grass systems utilizing organogenesis seem to be circumvented. Also, culture work is less labor-intensive because the embryos contain a complete plant axis and, in contrast to shoots produced via organogenesis, do not have to be further manipulated to induce development of other essential organs such as roots.

The selection of useful mutants can be facilitated because of the single cell origin of nonzygotic embryos. A mutation in the initial dividing cell would be carried by all cells of the developed embryo. Although there is a great deal of variability present in conventional embryogenic culture systems, the

potential for mass in vitro cloning of select genotypes exists and should become a reality once control of variability is achieved.

Finally, dormancy may be potentially induced in nonzygotic embryos. This would allow them to be processed to function as seeds with regard to efficient handling and dispersal.

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One of the functions of a speaker at a work session is to orient us in our topic and to provoke us to discussion. With that purpose, this contribution will use genetic and breeding studies with newly-cultivated forage legumes as examples of forage breeding objectives, and then as examples of some questions and problems about meeting those objectives. A better title might be "new examples of old questions".

Three of the best known genera of newly-cultivated forage legumes are Stylosanthes, Desmodium, and Leucaena. Macroptilium and Neonotonia are also fairly-well studied. All of these genera Aeschynomene, Alysicarpus, Arachis, Centrosema, Galactia, Lablab, Lotononis, Macrotyloma, Psoralea, Rhynchosia, Vigna, and Zornia are relatively little studied, beyond the performance of one or several promising collections. Some of them have been studied in cultivation for about 80 years, others for 10 years or less. Most activity has been in Australia and more recently in South America (Davies and Eyles, 1968; Hutton, 1970; Schultz-Kraft and Giacometti, 1978; Hutton, 1968; Cameron *et al.*, 1984, in press). Alysicarpus, Lablab, and D. heterocarpon are probably very old in cultivation in India but are new to European-heritage agriculture. Lespedeza is purposefully omitted because it is a unique topic in itself.

BREEDING OBJECTIVES

The first breeding objective for these genera is forage dry matter productivity. This passage from Hutton and Beall (1970) is a realistic statement about why we will breed for forage production when what is wanted is animal production. "Any improvement (in Siratro) should result in higher animal productivity from a pasture system based on this legume. However evaluation of new bred (M. atropurpureum) lines for their ability to increase animal production is dependent on the establishment of grazing trials when sufficient seed of the final selections is available. In the improvement program it has been necessary to assume that increased dry matter yield and other characters used as the basis for the selection will eventually result in increased animal production from this legume".

We emphasize forage dry matter because plants produce considerable stem and even wood dry

matter that is not eaten by cattle or sheep. Altered distribution of dry matter between leaves and stem is a breeding objective for Leucaena and Aeschynomene.

High dry matter production is a relevant character only after establishment and persistence are successful. Large seed size, seedling vigor, acetylene reduction, and early nodulating ability are related to each other and to ease of establishment in Desmodium (Hutton and Coote, 1972; Pinchbeck *et al.*, 1980). Field establishment can be affected by the ability of a seedling to nodulate effectively with native Rhizobia if inoculation fails. Rapid vegetative spread can be a critical component of establishment, for example, in Arachis glabrata, a very promising but largely sterile perennial.

Persistence also must usually be selected indirectly through component traits. Grazing tolerance and competitive ability are critical to persistence. Grazing tolerance and competitive ability are both complex traits and are often negatively correlated with each other. Legumes with twining and climbing growth habits such as Macrotyloma, Rhynchosia, and Centrosema compete very well with grasses but have less grazing tolerance than or stoloniferous prostrate types such as Lotononis or Stylosanthes humilis (Whiteman, 1980 pp. 138-157; 239-242). Breeding for more growing points per unit area may improve grazing tolerance and thus persistence of the twining legumes (Hutton, 1968).

Using indirect selection for component traits or using some other, even more rapid, form of genetic manipulation, it may be possible to achieve outstanding persistence potential in breeding material in a short time. Will there be a rapid way to test for persistence per se?

Increased palatability and/or the reduction of antinutritional compounds are a major breeding goal for some species, for example, for the highly productive, leafy and high-protein genus Indigofera (Hutton and Guerassimoff, 1966; Davies and Eyles, 1968; K. Quesenberry, pers. comm. 1984). Desmodium species contain from 1 to 11% tannin (Burnes and Forbes, 1962, cited in Rotar, 1965). This is about the same range of tannin as in sericea lespedeza (L. cuneata) (Henson, 1957). Tannin content may protect warm-season legume seedlings when a companion warm-season grass is young and highly palatable (Humphreys, 1982; Skerman, 1977, p. 122). It is reported that protein that is not digested in the rumen because it is complexed with tannin may in fact be digested and used more directly farther down the digestive tract; and tannin is associated with rare occurrence of bloat (Humphreys, 1982). If these benefits exist, then there may be an optimum of tannin content that we need to identify.

The economics of seed production can determine whether a valuable cultivar will be widely planted or not. This is the main reason that CIAT includes seed yield as a selection

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criterion for these new forage legumes (CIAT, 1983; Shultz-Kraft and Giacometti, 1978). For east of harvesting, seed should be non-shattering, ripen at one time, and be held above the leaf canopy. Determinacy and loss of the shattering trait are properties of domesticated plants and will need to be bred into newly-cultivated legumes. Lablab, Alysicarpus, and D. heterocarpon cv. 'Florida' show domesticated seed production traits. A. falcata, at the other extreme, produces shattering seed indeterminately beneath the canopy. These seed must be suction-harvested and are correspondingly expensive. They are however perfectly adapted for plant replacement within the sward.

If seed production is not possible or practical for some species, can these species be vegetatively propagated on a commercial scale? Vegetative propagation has the advantage that planting material is produced locally. Local production of seed or vegetative planting material avoids possible loss of adaptation to the livestock-producing area. Seed of the newly-cultivated legumes is being produced where used in Thailand (Humphreys, 1982) in Florida, and at CIAT (CIAT, 1983).

Breeders working with the newly-cultivated forage legumes are searching for variation that may extend ranges of adaptation. Four representative components of adaptation are daylength requirements, tolerance of sub-optimum soils, tolerance of climatic factors, and pest resistance. Many of these new species may have to be bred for earliness or photoperiod insensitivity. Valuable Desmodium and Stylosanthes species are short day plants (Rotar et al., 1967; t'Mannetje, 1965; Imrie, 1973) that are not induced to flower at our latitude before they are killed back by frost. When regrowth occurs in spring, the days are already too long to induce flowering. Variation for flowering time exists but early or photoperiod-insensitive genotypes would not be selected by evaluation programs based in the tropics because forage production generally declines as soon as flowering begins (Stobbs and Imrie, 1976). We will not be able to rely on germplasm evaluated and selected in the tropics.

Tolerance to acid soils will extend the range of a few species. CIAT has an active breeding program to improve the ability of Leucaena to grow in acid soils. Many warm-season legumes are acid-soil tolerant or even acid-soil indicator species (Schultz-Kraft and Giacometti, 1978).

Many of the most productive new forage legumes lack frost or drought tolerance. For example, within the species S. guianensis, genotypes of the tender botanical variety guianensis are productive in mixtures with vigorous grasses. These have been hybridized with genotypes of

the less productive but frost- and drought-tolerant botanical variety intermedia of the same species. This breeding effort has so far been unsuccessful because the F_1 's are largely sterile (Cameron, 1974; Cameron and Ludlow, 1977; t'Mannetje, 1977; Sumberg and Miles, 1982).

There are two ways that disease and insects can limit the useful range of introduced species. First, when a species is moved from one environment to another, it may encounter new wide-host-range pests to which it has no resistance. The species will then fail immediately. M. atropurpureum, a species of semi-arid habitats, fails in humid areas because of Rhizoctonia. CIAT is searching for Rhizoctonia-resistant genotypes for use in humid South America. Second, even if a species in a new environment is at first pest-free, problems will probably appear as the species is planted more extensively. For example, Stylosanthes is limited in its native South America by the co-adapted anthracnose disease fungus. Anthracnose has recently appeared in Australia. The use of single major genes for resistance may not be practical even if they exist because the evaluation period of a new forage cultivar is long and it may not be possible to introduce cultivars with new major genes fast enough. Breeders will have to use the potentially dangerous tactic of major-gene pyramiding or the time-consuming method of breeding for field (or horizontal) resistance (Cameron et al., 1984, in press).

SOME PERENNIAL QUESTIONS COMMON TO ALL FORAGE BREEDING

How can a sufficiently large number of genotypes be evaluated under grazing as early in the selection process as possible? Large numbers of entries can be evaluated for grazing tolerance with replication by short-term intensive controlled grazing (Hutton, 1962), but most of these entries must be discarded on the basis of component traits before it has been possible to do anything close to direct selection for animal performance. Under these restrictions, judgements must be made about entire species and about breeding materials within species.

If one selects indirectly for animal performance or persistence, or any trait y , the response one obtains will be a product of i , the selection intensity; the heritability of the trait x directly selected; the heritability of trait y itself; the genotypic correlation between x and y ; and the phenotypic standard deviation of trait y . This product is smaller by $(h_{r_{xy}})$ than the response if one could select for y directly (Bray, 1975). In order to choose trait x for the highest possible response in trait y requires specific genetic information: heritabilities of each trait and of y , correlations of each trait with y , and variance of y . For the newly-cultivated legumes in particular, this information rarely exists.

QUESTIONS WITH PARTICULAR REFERENCE TO THE NEWLY-CULTIVATED SPECIES

The first question that we confront is, "How to make an objective decision as to when to begin genetic studies and breeding? Should resources be concentrated in collection and evaluation of a genus or species until it is exhaustively known, to preclude breeding a cultivar that is inferior to or no better than the next introduction (Hutton, 1977)? If there are 50 legume genera containing 2,500 species worth investigating (Williams, 1964; cited in Kretschmer, 1967), can we wait to be certain that we have collected the best wild genotypes?

Siratro was bred when few collections were available (Hutton, 1962), yet in its area of adaptation it is superior to subsequent introductions. This is possibly the result of intelligent - or fortunate - plant collection. On the other hand, for the past twenty years, Australian literature has contained reports of breeding programs and superior hybrid progeny of various species almost ready for release (Hutton, 1968; Imrie, 1973; Gray, 1966; R. J. Clements, pers. comm., 1983). Out of all this breeding material, only one other cultivar, Leucaena leucocephala cv. 'Cunningham', has actually been released in Australia. Nothing else has been superior to the best collections. For some newly-cultivated species there is no or little well-characterized material from which to select parents. Dr. Harlan has a description for this quandry: he says "Every card in the deck is wild" (Harlan, 1976). Hutton (1968) and many others have suggested that the first crosses may be "shot-gun approach" crosses, made between unknown parents. These exploratory crosses should be made even if all accessions may appear similar and especially between accessions from different environments.

Eventually, however, plant breeding will be necessary for further improvement of any species. Plant breeding is needed because correlations between valuable characters are often negative. For example, typical pairs of negatively correlated traits are productivity and persistence, quality and disease resistance, productivity, and quality, and competitive ability and grazing tolerance. Seed yield and forage dry matter yield can be negatively correlated and earliness is negatively correlated with both. The improvement of one of a pair of negatively correlated traits with simultaneous gain or without loss in the other can only be accomplished by planned genetic manipulation. In fact, the management of conflicting selection objectives is the raison d'être of plant breeders.

Unlike most traditional temperate-climate forages that are cross-pollinated and self-incompatible, most of the newly-cultivated legumes are entirely self-pollinated. Some,

especially in Desmodium, are cross-pollinated but freely self-compatible. All of the traditional breeding methods and cultivar types associated with breeding self-pollinating or self-compatible species become available. Pure-line and single- or double-cross hybrid cultivars are genetically possible. What breeding methods will be used? What cultivar type (broad vs. narrow genetic base; heterozygous, heterogeneous, homozygous)? We need data on gene action, heritabilities, and variances in order to develop intelligent answers. Of the three bred cultivars that have been officially released, Cunningham Leucaena is from a single "F₄ line" (F₂-derived line in F₄) (Hutton and Beattie, 1976); Siratro is a bulk of three F₄ lines, that is, it has a broader genetic base; and a Hawaiian cultivar of Desmodium intortum, cv. 'Kuiaha', is a synthetic that is heterozygous and broader-based still (Rotar and Park, 1971; cited in Brewbaker and Rotar, 1982). None is a single-plant advanced generation selection.

Most species have perfect flowers and are cleistogamous. Those that are cross-pollinated are insect pollinated (Hutton, 1960; Rotar et al., 1967; Cameron et al., 1984 in press). These traits will make hybridization techniques laborious and the use of male sterility genes problematical. Cytoplasmic male sterility has been found in a spontaneous interspecific hybrid of Desmodium (McWhirter, 1969) but is has not been used to produce hybrid cultivars. Any type of recurrent selection and recombination will be costly to use with these species.

Few morphological or molecular marker genes are known and those that are available are generally useful only in intraspecific crosses (Imrie and Blogg, 1983; Hutton and Gray, 1967; Park and Rotar, 1978; Chow and Crowder, 1972, 1973, 1975; Robinson and Megarrity, 1975; Stace, 1982). Foreign pollen does not appear to have an advantage, at least in Desmodium (Chow and Crowder, 1973). Frequent selfs occur during controlled pollination (Chow and Crowder 1973) and these are not readily identified. How can crosses be quickly and reliably distinguished from selfs?

Present taxonomic distinctions provide little guidance for identifying genetic distance or similarity. In Stylosanthes, fertility barriers exist within the species S. guianensis. By contrast, in Desmodium there is a species, D. sandwicense, that is disallowed by some taxonomists (J. Zarucchi, Mo. Botanical Garden, pers. comm., 1984) who include it in D. uncinatum. Yet D. sandwicense will hybridize and introgress spontaneously with D. intortum while hybrids of D. uncinatum with D. intortum show a high degree of sterility (Hutton and Gray, 1967; Stobbs and Imrie, 1976; Chow and Crowder, 1972, 1973). As these legumes have become better known, taxonomic revisions have become necessary and frequent and communication sometimes breaks down entirely.

As soon as we think about working with legumes, we think about breeding for increased nitrogen

fixation. However, given that successful symbiosis is probably necessary for persistence and productivity of these new legumes and that any symbiotic N-fixation in the warm-season is better for the pasture system than none, what priority should be given to breeding for high levels of N-fixation per se? Is it worth diverting resources from other breeding objectives?

To sum up, forage dry matter productivity, reliable establishment, and persistence are, world-wide, the primary breeding objectives for the newly-cultivated legumes. Other breeding objectives are economic yields of harvestable seed or vegetative propagules and maximum use of variation that may extend the range of adaptation of these new species.

Some of the problems of breeding the newly-cultivated legumes are common to all forage breeding. Most of the variation for effect on animal performance within a species of population can be evaluated indirectly only. For practical reasons, indirect selection is unavoidable yet it is genetically inefficient.

Some questions are unique to newly-cultivated species. Adequate evaluation of the germplasm resources is an essential and resource-consuming task. At what stage in germplasm collection and evaluation should breeding begin? What breeding methods and cultivar types should be used? There is little data yet available for making informed breeding decisions.

Genetic and breeding studies with newly-cultivated forage legumes bring up old questions and create a few new ones. Every forage breeder is familiar with most of these situations. Each breeder will have a working perspective on these questions that is derived from their own experiences with forage crop improvement.

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SYSTEMATIC COMPARISONS AMONG ZEA MITOCHONDRIAL DNAs

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Three independent genetic systems are now recognized in higher plants, the nuclear, chloroplast (ct), and mitochondrial (mt). The later two manifest cytoplasmic or extra-nuclear inheritance, the basis of which is the presence of unique ctDNA and mtDNA molecules. Inheritance of the organelles is essentially uniparental; that of the nucleus is biparental. Clearly, these two systems produce rather distinct patterns of diversity.

The patterns of organelle diversity and the interrelationships of the uniparental and biparental systems are of fundamental importance to a) understanding the evolution and systematics of the organelle DNA, b) assessing and cataloging of cytoplasmic variability for more efficient utilization of diverse germplasm, and c) providing an effective strategy to combat the hazards of genetic vulnerability at the cytoplasmic level.

We analyzed restriction endonuclease fragment patterns of ct and mtDNAs from six races of teosinte and a normal US corn belt-type maize (Timothy *et al.* 1979). The analyses of ct DNAs from the eight teosinte collections revealed three different groups: a) Perennial teosinte and Guatemala; b) Huehuetenango and Balsas; c) Chalco, Central Plateau, Nobogame, and El Salado (a sample of Balsas). Mitochondrial DNAs were placed in four groups by restriction endonuclease analyses: a) Perennial teosinte; b) Guatemala; c) Nobogame; d) all other teosintes. These results were most significant in that they formed a basis from which to begin formulating concepts of organelle evolution in higher plants: 1) Diversity exists among organelle DNAs of a species complex, 2) mitochondrial and chloroplast DNA systems may vary independently of each other, and 3) organelle DNAs do not appear to evolve independently of the nuclear system, but evolve in reasonable concert with the nuclear system. An important facet of this work is the demonstrated feasibility of comparative analyses of a hierarchy established by organelle DNA differences versus the relationship established by biosystematic classifications in terms of the total organism (conventional systematic determinations by taxonomic, genetic and cytogenetic methods).

The mitochondrial genome of maize may be as large as 320 megadaltons (Ward *et al.* 1981). It is isolated as an array of linear DNA

molecules or covalently closed circular molecules. The frequencies of the variously sized circular molecules in the mtDNA complement varies among the fertile (N) and S and T forms of cytoplasmic male sterility (cms) (Levings *et al.* 1979).

The cms-T, -C, and -S mtDNAs have restriction endonuclease digestion patterns that are distinctive from each other and from those of N cytoplasm. The cms-S is further distinguished by the presence of two unique plasmid-like molecules, S-1, and S-2 (Pring *et al.* 1977). These molecules are 6.4 kilobases (kb) and 5453 base pair (bp), respectively (Levings and Sederoff, 1983), with each molecule having identical terminal inverted repeats of 208 bp. The cms-S occasionally reverts to fertility, with the disappearance of the plasmid-like molecules and their apparent integration into differing regions of the main portion of the mitochondrial genome (Levings *et al.* 1980). These findings are in accord with transpositional events.

Another unique set of plasmid-like DNAs was found among fertile South American maize races (Weissinger *et al.* 1982). These plasmids, found in RU cytoplasm, were in 17 of 93 races from Latin America (Weissinger *et al.* 1983), are R-1 and R-2. Yet another set of plasmids designated D-1 and D-2 were found in ZD cytoplasm from diploperennial teosinte (Timothy *et al.* 1983). R-1 and D-1 are about 7.5 kb with terminal inverted repeated sequences of about 0.2 kb. R-2 and D-2 are about 5.5 kb in length with a terminal inverted repeat of 0.2 kb. No differences have been found among D-2, R-2 or S-2 molecules. The main feature that differentiates the D-1, R-1, and S-1 molecules is a 1.5 kb sequence found in S-1, and also contained in S-2, but not found in R-1 or D-1. No dissimilarity has been found between the D-1 and R-1 molecules. The S-1 molecule might have originated from a recombinational event between the R-1 and R-2 DNAs (Levings *et al.* 1982)) or from a similar event between D-1 and D-2 (Timothy *et al.* 1983). The natural occurrence of cms-S thus far appears restricted to Meso- and North America, whereas the RU and ZD cytoplasms are essentially South and Meso-American. Unlike cms, the RU and ZD cytoplasms are not associated with cytoplasmic male sterility or any other known phenotypic trait.

It has become increasingly apparent that variation of organelle DNA within the species complex of the organism may be the common condition. From 93 accessions of Latin American maize races we have obtained ten BamHI fragment patterns and eight from EcoRI (Weissinger *et al.* 1983). Races were placed into one of eighteen groups, each group being defined by a particular combination of BamHI and EcoRI patterns and the presence or absence of plasmid-like DNAs. Seven races produced patterns identical to those produced by one or another of the major cms groups (C, T and S) with each group being represented by at least

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one race. Fertile races from Meso-America and some South American races with Meso-American affinities were separated from other South American races. South American races were divided among three general classes of related groups. There was considerable agreement between these groupings, and those derived by other workers on the basis of morphological and cytological affinities. This and our other work form the basis upon which a catalog of classes of cytoplasmic variation can be made.

If the plasmid-like DNAs behave as transposable elements, their effects on sequence rearrangements are not yet understood. However, six races of maize having the R-1 and R-2 plasmid-like DNAs were found to have, when digested by BamHI and EcoRI, five distinct fragment patterns. These digestion patterns were different from the standard RU fragment patterns exhibited by the twelve races comprising that group (Weissinger et al. 1983). Similarly, digestion patterns of 16 stocks of diploperennial teosinte cytoplasm (2D) containing the D-1 and D-2 molecules, revealed two disparate classes with minor variation within each. Differences of this magnitude occur at the specific or subspecific level, but have not been seen at the varietal or racial category. Consequently, the structural organization of the genomes in these two major classes must differ greatly.

Differences in sequence may also occur by simple base changes or substitutions. However, evolution of the mitochondrial genomes in Zea has occurred with major rearrangements of sequences, although there is general sequence conservation (Sederoff et al., 1981). Intricate and complex changes in homology and position of fragment patterns are best explained by duplications, deletions, transpositions, translocations, or inversions.

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ASSOCIATIVE N₂ FIXATION--AN UPDATE

Rex L. Smith¹ and S. C. Schank¹

The discovery of the N₂-fixing association of grass roots with Spirillum lipoferum (since classified as Azospirillum brasilense) by Dr. J. Dobereiner and co-workers in Brazil was reported in 1974. In that report, two most important observations were reported: 1) Root tissues seemed to be infected with the N₂-fixing bacteria, and 2) N₂-fixing bacteria, associated with grasses, could fix economically important amounts of nitrogen. The implications of the existence of an efficient N₂-fixing system in grasses could fix a significant portion of the nitrogen they require for growth. Research to define and develop associative N₂-fixing systems was initiated and has continued in numerous laboratories worldwide. The purpose of this paper is to update the concepts that research has elucidated and to assess the potential as well as the problems of those systems.

NATURE OF THE ASSOCIATION

Early observations, using light microscopy and tetrazolium staining, indicated that Azospirillum could infect grass root tissues (Dobereiner et al 1974). We made observations using light microscopy and specific immunofluorescence labeling of Azospirillum where the bacteria appeared to be inside root cortex cells (Schank, S. C. and R. L. Smith, unpublished). However, that method did not have sufficient resolution to verify whether or not those root cells were living, or to reveal details of intracellular interaction. In subsequent observations of samples from inoculated field plots, N₂-fixing bacteria were observed mainly in the rhizosphere and on the rhizoplane (Schank et al 1979). Similar data has been collected using scanning and transmission electron microscopy (Schank et al 1983). Matthews et al (1983) used peroxidase-antiperoxidase staining in conjunction with electron microscopy to show the rhizosphere-rhizoplane relationship. Our experience has always shown that N₂-fixing bacteria are on or near the root surface or in non-living cortical cells. Umali-Garcia (1980) reported similar results, but with evidence that root cortex tissue could be invaded intercellularly. We agree with Patriquin et al (1983) that further studies are required to confirm whether Azospirillum invades the inner root regions.

The closeness of the root-bacteria association is directly related to the efficiency of associative N₂ fixation. Internal infection could provide a closed system which could efficiently couple energy transfer from the

plant to the N₂-fixing bacteria, transfer fixed N from the bacteria to the plant, and possibly provide a mechanism to protect nitrogenase from inactivation by excessive O₂. As it now stands, only loose bacteria-plant associations are believed to operate which allow competition among soil organisms for energy and fixed N with concomitant low N₂-fixing efficiency.

GRASS RESPONSES TO INOCULATION WITH AZOSPIRILLUM

The desire to manage associative N₂-fixation systems prompted early inoculation experiments in Florida. Results of those studies were encouraging as significant dry matter yield increases (up to 80%) due to inoculation were observed (Smith et al 1976, 1977). Extrapolations indicated that inoculation stimulated yield responses equivalent to application of an additional 30-40 kg N/ha. However, it soon became apparent that the yield responses to inoculation were erratic and about half the time significant yield responses were not obtained.

Inoculation experiments have been conducted at many locations around the world since 1974. Most of those areas have reported economically important but inconsistent yield responses. One exception has been Israel where numerous inoculation experiments with wheat, corn and other grasses have produced consistent yield responses (Cohen et al 1980 and Kapulnik et al 1981). The Israeli trials were on fine textured, alkaline soils and were well irrigated. Researchers, however, attributed part of their success to inoculation with Azospirillum brasilense strain Cd. Following the Israeli successes, we conducted five experiments on three different sites using the Cd strain (Smith et al 1984). Four experiments on two sites were conducted in Florida. The other experiment was in New Mexico because its soil and climate resembled Israel's. Only two of the Florida experiments gave positive inoculation responses while the New Mexico site gave none. This less than 50% success rate showed that strain Cd is not universally successful and, in fact, did not perform better than strains used earlier.

Research into the effects of management factors has given some insight into improving response consistency. Moderate rates of nitrogen fertilization (Smith et al 1976) promote inoculation response. Plant genotypes (Bouton et al 1979) and bacterial strains (Schank et al 1984) vary in their ability to produce inoculation responses. Application of the best known technology still gives a level of response consistency unacceptable for commercial recommendation in Florida. However, commercial inoculation with Azospirillum is being planned in Israel.

Nitrogenase activity in our research plots has not been correlated with inoculation treatment (Smith et al 1984), nor has nitrogenase activity been associated with yield responses (Smith et

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al 1978, 1984). Both of the two positively responding Cd inoculated experiments had non-significant N_2 fixation rates indicating that N_2 fixation is not a necessary component for yield responses to inoculation.

Azospirillum has been shown to produce compounds with phytohormone activity capable of causing root enhancement (Tien et al 1979). Lin et al (1983) reported that root elongation and branching in field- and pot-grown grasses have been increased by inoculation with moderate Azospirillum numbers, whereas, heavy inoculation rates caused inhibition of root development. Inoculation also improves mineral uptake. Uptake of NO_3^- , K- and $H_2P_2O_4$ by corn seedlings was enhanced 30-50% over controls. Increased mineral uptake could account for the inoculation yield responses.

N_2 FIXATION IN ASSOCIATIVE SYSTEMS

Initial claims of very high rates of N_2 fixation (Von Bulow and Dobereiner 1975), nearly equal to that of legumes, have not been verified and it is generally believed that the methods used in those reports artificially inflated the data. However, several laboratories have shown that N_2 fixation does occur and have verified it by the incorporation of $^{15}N_2$ into plant tissue (Fuschel et al 1975). Evaluation of the potential fixation rates of associative N_2 fixation has been difficult and has given only rough estimates. Improved acetylene reduction (AR) assay methods are now used extensively to measure associative N_2 -fixation but even those methods have serious limitations. Soil-root cores and in situ AR assay methods are preferred, but at best, the data is highly variable and should be interpreted with caution.

From AR activity measurements on soil-root cores, we estimate that maximum rates of fixation are about 20-30 kg N/ha (Smith et al 1982). In Florida, most sites have low AR activities with insignificant rates of nitrogen being fixed, especially in well-drained, sandy soils. Several high AR activity sites have been found (Smith et al 1982) and high activity has always been associated with high soil moisture. We believe that a major effect of the high moisture is to reduce the O_2 in the zone of N_2 fixation in porous soils.

Matched core comparisons, of cores containing living grass roots compared to root-free cores, showed that much of the nitrogenase activity was plant dependent (Schank and Smith 1984). Root-containing cores had about 10-15 fold higher activity than soil alone, providing strong evidence for the associative nature of N_2 fixation. Plant maturity and condition also affected nitrogenase activity. Nitrogenase activity increased over time, with plant development reaching a maximum during anthesis and early heading (Smith and Schank

1980). Depletion of root energy reserves following clipping (Schank et al 1984) and reduced solar radiation by shading (Smith et al 1984) also decreased nitrogenase activity.

We have found that bacterial inoculation has little or no impact on AR activity (Smith and Schank 1980). The measured activity appears to be mainly from indigenous N_2 -fixing bacteria which are always present but not always active.

FATE OF INOCULATED AZOSPIRILLUM IN THE SOIL

Good inoculum survival and colonization are necessary to have positive impact in N_2 -fixation and stimulate yield increases. Very little data is available on the fate of the inoculated organism following inoculation. The main problem in collecting meaningful data is positive identification of the inoculum strain in the counting procedure. Much of the information in the literature is questionable because non-specific identification criteria were used.

Using fluorescent antibody techniques, we found that inoculated Azospirillum strains survived from year to year but in low numbers (Schank et al 1979). Mutant strains of A. brasilense 13tSR and CdSR, resistant to streptomycin and rifampin, were isolated to facilitate their identification and recovery in colonization studies. In addition, strain Cd produces a unique reddish pigmentation under certain conditions that permits positive identification. In field colonization studies, we found that the inoculum population declined rapidly after inoculation. By the sixth week after inoculation, Azospirillum populations had dropped from about 10,000 per g of soil to only about 100 (Smith et al 1984).

Studies were made to determine what factors affect survival of inoculated Azospirillum (Albrecht et al 1984). We found high organic matter and high moisture (tested to 90% of F.C.) promoted survival and that soil gave better survival than sand. In pre-sterilized systems Azospirillum numbers remained relatively stable for several weeks while in non-sterilized systems bacterial numbers dropped rapidly. Indigenous bacterial populations, monitored as controls, remained stable. This information tells us that in our Florida soils, biological factors are the main deterrents to maintaining inoculum numbers.

We consider poor inoculum survival and colonization to be the most serious problem in inoculation technology. Those problems may be responsible for inconsistent yield responses and failure of inoculation to affect nitrogenase activity. Research on that problem should be given top priority.

FUTURE OUTLOOK

Nitrogen fixation potential is severely limited

by: 1) inefficient transfer of energy from the plant, 2) inefficient transfer of fixed nitrogen back to the plant, 3) inadequate nitrogenase protection from excessive O_2 and, 4) poor inoculum survival. Those problems will not be easily solved as they hinge upon the loose nature of the plant-bacteria association which does not lend itself to manipulation. However, some sites exist that sustain significant natural N_2 fixation. Biotechnological research directed toward increasing the competitive ability of N_2 -fixers in the rhizosphere, broadening N_2 -fixer substrate utilization to include plant structural carbohydrates, and improving O_2 protection mechanisms could make real increases in the N_2 -fixation potential.

Economically important enhancement of yields by inoculation is attainable in many grass crops but inconsistency is the main problem. We now believe that those yield responses are not due to N_2 fixation and research methods should be modified accordingly. Without N_2 fixation, the enhanced yields could either reduce the soil fertility or improve plant nitrogen use efficiency. If yields can be enhanced without reducing soil fertility, then inoculation may be valuable. However, if the improved yields are at the expense of soil fertility, the long term value of inoculating grass crops may be nullified. It is important that this issue be resolved.

Improvement of inoculum survival and root colonization may be key in improving the inoculation response consistency and warrants research. Genetically engineered Azospirillum that over produce phytohormones are now available and may be of value in both improving consistency and resolving whether growth responses are due to phytohormonal activity.

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UPDATE ON FESCUE TOXICITY RESEARCH AND RELATED MATTERS

Donald M. Ball¹

I believe most of you are aware of the fact that Auburn University recently released a new tall fescue variety names 'AU Triumph'. This variety has yielded well in Alabama variety trials and has been shown to be more compatible with legumes than 'Kentucky 31'. However, its primary attribute is that it makes more winter growth than other varieties currently commercially available. We feel that this is a valuable characteristic because it can help cattlemen reduce their stored feed requirements.

We expect 'AU Triumph' will be commercially available in Fall, 1984. It is probable that several hundred thousand pounds of seed will be sold by the end of 1984, although most of this will be marketed within Alabama. It should be emphasized that all of this seed will be free of the fungus Acremonium coenophialum which has been implicated in fescue toxicity. If you have an experiment or a demonstration in which you would like to include 'AU Triumph', I feel sure it will be possible for you to obtain seed in Fall, 1984.

I also would like to give you a brief report on the Auburn University Fescue Toxicity Diagnostic Center. This facility was opened to the public June 1, 1983 and was established for the purpose of giving livestock producers and seedsmen a place to have either plant tissue samples or seed of fescue tested for the Acremonium fungus. This was the first laboratory set up primarily for the purpose of producer service, and I'm happy to report that it has apparently been very well received by producers. As of March 31, 1984, a total of 1725 samples had been tested in the Fescue Toxicity Diagnostic Center. The majority of these had been submitted by producers, although the figure includes some samples analyzed for research purposes.

The fact that samples have been received from 21 different states as well as two foreign countries is indicative of widespread interest in this topic. It is interesting to note that the mean infection level for samples submitted has been around 70% (meaning that on the average, around 70 out of each 100 plants or seed in the pastures or seed lots the samples represented were infected). However, it is extremely important to note that the range of infection has varied from 0 to 100%. Therefore, a producer cannot make accurate assumptions regarding the level of infection he has in his pastures. The only way to know what level of infection, if any, exists is to have them tested.

As you know, there has been a great deal of

research in the fescue toxicity area in recent years and there have been many papers presented on this topic. Much work needs to be done

before we fully understand fescue toxicity, but it now seems clear that the endophytic fungus is involved in, if not totally responsible for, causing this problem. I would like to take a few minutes to summarize what I see as being the logical options which producers currently have regarding taking advantage of the information which research has made available to date.

I believe it is fairly obvious that if a producer is going to establish a new planting of fescue which will be used for grazing or hay, he needs to be certain that he is using fungus-free seed. Fungus-free seed of a number of fescue varieties is currently available, and the quantity of commercially available fungus-free seed should increase rapidly in the near future. The relationship of supply and demand should insure that producers can purchase fungus-free seed at reasonable prices.

Next the question arises, "What options are available for a producer who has existing stands of fescue?" In my mind, the first logical step would be for a producer to determine the "fungus status" of his existing pastures. He can do this by taking samples at random from throughout a field and having them tested at the Auburn University Fescue Toxicity Diagnostic Center, or at another lab which is offering this service.

It is probable that a producer will find that most of his existing pastures will contain some level of fungus infection. Obviously, if there are varying levels of infection in different pastures, he would be well advised to put his first efforts on reducing the effects of the fungus in those pastures which are most heavily infected.

There are three options which producers currently have for reducing the effects of the fescue fungus. The first of these is to grow legumes with the fescue, the two most logical species to use in most situations in the Southeastern United States being white clover or red clover. Research conducted at Auburn University has indicated that having as much as 20-25% of an infected fescue stand being comprised of legumes will go a long way toward offsetting the effects of the fungus. A second option would be to dilute the amount of fungus-infected fescue which animals are receiving. As an example, if an animal is getting almost 100% of its nutrition from fungus-infected fescue forage and a producer begins feeding some other materials such that the animal is then getting only 50% of its nutrition from fungus-infected forage, then the adverse effects of the fungus will likely be greatly reduced.

The third option would be for a producer to start over by reestablishing a pasture which is infected. If a producer decides to do this, he should prevent the field from producing seed during the year that he reestablishes it. This

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is for the purpose of reducing the amount of seed in the pasture which might result in establishment of infected volunteer plants.

The existing plants in an infected pasture which is to be reestablished will, of course, have to be killed. A producer could do this either through tillage, chemicals, or a combination of the two. It appears that paraquat would be an effective and relatively economical chemical to use to kill established infected plants. After the plants have been killed, the producer would then simply need to replant using fungus-free seed. Experiments conducted to date give every indication that pastures which are reestablished in this manner will be fungus-free and will remain fungus-free.

A very interesting and important question is, "What level of fungus infection can a livestock producer tolerate?" The truth of the matter is that we really don't have enough information at present to give a good answer to this question. However, in a four year study with steers at the Black Belt Substation in West Alabama, animals grazing fungus-free fescue produced 125 pounds more beef per acre than animals grazing fungus-infected fescue. The value of this additional gain is enough to make it very attractive to a producer to reestablish a highly infected pasture.

Data obtained at Auburn University indicate that young animal gains seem to be lowered by between 0.1 and 0.2 pounds per day for each 10% of fungus infection. I think you will agree that this level of reduced performance is very economically important. There are a number of factors which a producer may need to consider prior to making a decision as to whether or not to reestablish a particular pasture including, percent infection, percent clover in the pasture, percent fescue in the animal's diet, cash flow, and crop rotation options. The "bottom line", however, is that many producers can justify reestablishing an existing infected pasture.

The potential economic impact which could result from using fungus-free, rather than fungus-infected fescue is tremendous. I mentioned earlier that in a four year study at the Black Belt Substation, steer gains were increased by 125 pounds per acre. Also, surveys have shown that well over 90% of existing fescue pastures in Alabama are infected. If one assumes only a 50 pound per acre per year increase in performance, and if beef is worth 60¢ a pound, this means the potential increased value of production from the 850,000 acres of fescue in Alabama is over 25 million dollars annually! If this applies to all fescue in the nation and if we use these same assumptions in calculating the increased value of production by using fungus-free rather than fungus-infected fescue on the 35 million acres of fescue in the United States, the potential increase value is over 1 billion dollars annually! It should be

emphasized that these figures do not include any estimates of value obtained by increasing reproductive performance of cattle. Research in progress is beginning to indicate that the fungus may be having a very detrimental effect in this regard.

I believe that the information obtained from the fescue toxicity research which has been conducted in recent years constitutes one of the most important developments in the forage/livestock industry in history. Even though we have had problems with fescue toxicity, producers have seen fit to establish some 35 million acres of this species in the United States since the release of 'Kentucky 31' in 1943. At present, fescue is in the spotlight more than ever. Most of our scientific advancements come in small increments, but in this instance we are seeing a major increase in animal performance on a very important and widely-grown forage crop. I believe that those of us who work as Extension Forage Crop Agronomists in states where there is a substantial acreage of fescue will be remiss if we do not place a great deal of emphasis on this topic in our Extension educational programs.

GRAZING LANDS MEETINGS AND GROUPS

Lamar Kimbrough¹

GRAZING LANDS AND PEOPLE NATIONAL LEADERS CONFERENCE

This group met on July 14-16, 1982 in Denver, Colorado. The Extension Service co-hosted this conference with the National Cattlemen's Association. A main idea was to increase public support for the proper management of all grazing lands.

Several needs identified at this meeting were:

- 1) Enhance use of grazing land products to meet human needs.
- 2) Establish understanding of economic values of grazing lands.
- 3) Develop awareness of non-commodity values and inter-relationships.
- 4) Identify cultural/heritage values of grazing lands.
- 5) Build recognition/understanding of the characteristics of grazing lands.
- 6) Promote understanding of stewardship responsibilities.

GRAZING LANDS TASK FORCE

In response to the above listed needs, ECOP approved the creation of an Ad Hoc Grazing Lands Task Force. This group met in Denver, Colorado on September 18-20, 1983. The major responsibility of the Task Force has been to develop an Extension Plan (strategy) for programming which will address the needs from the Grazing Lands Conference and to assist in implementing the same. The Task Force will seek ways for developing effective program relationships among organizations which are concerned about grazing lands and the Cooperative Extension Service.

The target audiences include policy makers, special interest groups, grazing land managers, service and support groups, youth, consumers, and urban publics.

GRAZING LANDS FORUM

The need for a Grazing Lands Forum was identified and its formation encouraged by a 1981 International Grasslands Congress resolution and the 1982 National Conference on Grazing Lands and People. The Forum plans to develop effective program relationships among organizations which are concerned about grazing lands. For more information concerning the Forum contact Evert Byington, the Executive Secretary, Winrock International, Petit Jean Mountain, Morrilton, Arkansas 72110, phone (501) 727-5435.

NATIONAL EXTENSION WORKSHOP

ECOP has approved this workshop to concentrate on the Management and Educational Programs for Grazing Land Resources. It is to be held at Denver, Colorado on September 24-27, 1984. The objectives of the workshop are:

- 1) To increase the level and effectiveness of communications among CES personnel, other interested agencies and organizations and the various states.
- 2) To increase the understanding of Extension's opportunities for increased CES educational programming for grazing lands.
- 3) To develop mechanisms for regional networking among CES subject matter specialists to facilitate the sharing of Extension materials, programs and ideas.
- 4) To develop strategies to obtain appropriate recognition of CES programs in grazing land education and management.
- 5) To identify and develop leadership among CES specialists to assist in coordinating education, conservation and management activities among their colleagues in the Land Grant Colleges, federal agencies and other groups.

Prime audiences are Extension Specialists in Range, Pasture, and Forage and appropriate Natural Resource Program Leaders.

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EFFECT OF ENDOMYCORRHIZAE ON GROWTH AND NITROGEN FIXATION OF CLOVERS

J. B. Mott¹ and D. A. Zuberer²

Extensive land disturbance is occurring in Texas as a result of surface mining for lignite deposits. Studies have shown that most of the mine spoils are deficient in both nitrogen and phosphorus (Dixon et al., 1980). Thus, fertilization during reclamation is essential to establish good vegetative cover. The use of legumes, principally clovers, in land reclamation and forage production is becoming an established practice to reduce the requirement for nitrogen fertilizer. In addition to symbiotic associations with Rhizobium, legume roots may be infected with endomycorrhizal fungi, thus forming a tripartite association. The plant receives fixed nitrogen from one symbiosis and benefits from enhanced nutrient uptake, especially of phosphorus, from the soil via the endomycorrhizal fungal hyphae (Mosse et al., 1981).

After initial field work showed that plants growing in mine spoils were able to form symbiotic associations with Rhizobium and endomycorrhizal fungi (Mott, 1984), growth chamber studies were conducted to investigate the effects of these two symbiotic associations on subterranean clover (Trifolium subterraneum L) grown in mine spoil at different fertility levels. Control plants were not inoculated, while treatments included inoculation with R. trifolii (Nitragin Co.), mixed mycorrhizal fungi in native topsoil taken from the mine area, or a combination of both R. trifolii and mycorrhizal fungi. Additional controls received a water soluble extract of the mycorrhizal inoculum and plants were also grown in unmined soil for comparison.

Growth in spoil was apparently limited by both nitrogen and phosphorus, and in unfertilized spoil only plants infected with both mycorrhizal fungi and R. trifolii responded with increased dry matter production (approximately twice that of any other treatment), as shown in Table 1.

Table 1. Yields (shoot dry weights) of subterranean clover inoculated with R. trifolii, mycorrhizal fungi, or a combination of both, grown in mine spoil.

Treatment	Unfertilized	Fertilizer*Fertilizer level 1 level 2	
			-----mg/plant-----
Uninoculated	34.5 h#	64.1 fg	175.5 c
Mycorrhizae	39.4 h	68.1 ef	182.8 c
<u>R. trifolii</u>	44.7 gh	102.1 d	304.9 b
Mycorrhizae + <u>R. trifolii</u>	90.0 de	177.7 c	390.7 a

*Level 1 = 19 kg N/ha, 8 kg P/ha; level 2 = 57 kg N/ha, 25 kg P/ha.

#Means of each variable followed by the same letter or letters are not significantly different at the 0.05 probability level by Duncan's Multiple Range Test.

Interaction of the two symbioses was evident at each level of fertility with dually infected plants in fertilized spoil producing 28-74% more shoot biomass than equivalent plants inoculated only with R. trifolii. Shoot dry weights of dually infected plants in unfertilized spoil were as high as those inoculated only with R. trifolii, grown in spoil at fertilizer level 1 (19 kg/ha; 8 kg P/ha).

There was no increase in growth as a result of inoculation with endomycorrhizal fungi alone, at any fertility level. However, there was a strong correlation between shoot phosphorus concentration and mycorrhizal infection ($r=0.63$; $P=0.0001$), confirming the role of endomycorrhizal fungi in enhancing uptake and translocation of phosphorus.

Dually infected plants exhibited significantly increased root growth, leaf number, nitrogen fixation, and nitrogen and phosphorus contents of shoots in comparison with plants infected with either microsymbiont alone. Plants inoculated only with R. trifolii failed to show increases in nitrogen fixation (acetylene reduction activity) when compared with control plants in unfertilized spoil (Table 2). In contrast, dually inoculated plants exhibited significantly greater activity showing a synergistic effect of endomycorrhizal fungi on root nodule nitrogenase activity probably as a result of improved phosphorus nutrition and plant growth as suggested by Smith et al. (1979).

In fertilized spoil, dually inoculated plants also showed higher acetylene reduction activity than those inoculated only with R. trifolii, with activity by these plants in lower fertility spoil equivalent to that of plants inoculated only with R. trifolii grown at a higher fertility level.

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Table 2. Nitrogen fixation (acetylene reduction activity) of subterranean clover inoculated with R. trifolii mycorrhizal fungi, or a combination of both, grown in mine spoil.

Treatment	Unfertilized	Fertilizer* level 1	Fertilizer level 2
	-----nmol C ₂ H ₄ plant ⁻¹ hr ⁻¹ -----		
Uninoculated	16.2 d#	98.6 d	114.5 d
Mycorrhizae	48.3 d	69.3 d	435.3 c
<u>R. trifolii</u>	145.6 d	325.0 c	813.3 b
Mycorrhizae + <u>R. trifolii</u>	375.7 d	732.5 b	1306.0 a

*Level 1 = 19 kg N/ha, 8 kg P/ha; level 2 = 57 kg N/ha, 25 kg P/ha.

#Means of each variable followed by the same letter or letters are not significantly different at the 0.05 probability level by Duncan's Multiple Range Test.

In fertilized spoil, dually inoculated plants also showed higher acetylene reduction activity than those inoculated only with R. trifolii, with activity by these plants in lower fertility spoil equivalent to that of plants inoculated only with R. trifolii grown at a higher fertility level.

Neither symbiosis was inhibited by the addition of 57 kg N/ha plus 25 kg P/ha, and in a second experiment which included a higher fertilizer treatment, only the mycorrhizal association was inhibited by the addition of 114 kg N/ha plus 50 kg P/ha.

Our data suggests that use of plant-microbe symbioses which can reduce fertilizer requirements could be of great significance in reducing costs of mine spoil reclamation and developing more self-sustaining revegetation techniques. The lack of inhibition of the symbioses at fertilizer levels used in reclamation (fertilizer level 2) also implies that these symbioses could be utilized to improve growth and nitrogen fixation of plants in current revegetation programs. However, further greenhouse and field studies are needed to assess the extent of the symbiotic advantage and fertilizer requirements of dually infected plants.

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THE INFLUENCE OF INOCULATION OF LEGUME ESTABLISHMENT

L. A. Materon¹ and R. W. Weaver¹

Successful establishment of forage legumes requires the seed to be inoculated at planting with the appropriate strain of *Rhizobium*. Failure of inoculation, absence of specific bacteria, and ineffective indigenous rhizobial populations will result in drastic yield reductions. Environmental factors (soil temperature, moisture, pH, and fertility), host-*Rhizobium* interactions (compatibility, strain infectiveness and effectiveness), as well as inoculation techniques markedly affect the legume response to nodulation by the inoculant strains (Giddens *et al.* 1982). In addition, the establishment of forage legumes depends largely upon the development of a functional N₂-fixing symbiosis between the legume and the specific microsymbiont. Thus, the introduced rhizobia should be effective in fixing nitrogen as well as capable of competing for nodule sites with the naturalized populations in the soil.

Seeding before the onset of rain may significantly reduce the number of seed-applied rhizobia especially if dry conditions persist. Survival of rhizobia on seeds can be extended by the use of proper inoculation techniques (Date, 1968, Giddens *et al.* 1982, Rich *et al.* 1983). Also, it is important to sow when the soil moisture is adequate for germination.

The presence of toxic factors on the seed of subclover and arrowleaf clover may decrease the multiplication of the seed-applied rhizobia and reduce the chances of nodulating the seedling. Seed-coats release a soluble toxic substance(s) that inhibits the growth of rhizobia when cultured in yeast-extract mannitol agar plates (Materon and Weaver 1984). Normal inoculant components such as peat, gum arabic and sucrose did not neutralize the toxins but extended survival of the rhizobia by protecting against desiccation (Materon and Weaver 1984) (Table 1).

Table 1. Effect of individual inoculant components and phenolic adsorbents on the growth of *R. trifolii* around Amclo arrowleaf clover seeds on the surface of yeast-extract mannitol agar plates.

Seed Coating +	Inhibition Diameter ‡ (mm)
Non-coated	7.9a*
Sucrose	7.8a
Peat	7.0ab
Pelgel	6.9ab
Gum arabic	6.2b
Activated charcoal	1.1c
Polyvinylpyrrolidone	1.2c

*Means not sharing the same letter differ significantly (P = 0.05) according to Duncan's Multiple Range Test.

+Water was used as the adhesive for peat, charcoal and polyvinylpyrrolidone (PVP).

‡Mean value from three strains of *Rhizobium trifolii* using 40 seeds per strain.

Using normally-inoculated seed coated with phenolic adsorbents, such as charcoal and polyvinylpyrrolidone (PVP) partially neutralized the toxins and greatly increased the survival of the seed-applied rhizobia (Table 2). However, some seed from a selection of arrowleaf clover cv 'Amclo' provided by Dr. Ray Smith at the Texas A&M University Agricultural Research and Extension Center at Overton, were found non-toxic to the growth of rhizobia and therefore, supported more rhizobia than the amended toxic seed of arrowleaf clover after inoculation (Table 2).

Table 2. Survival of *Rhizobium trifolii* (strain 162Y10) on toxic arrowleaf clover seed as affected by the incorporation of phenolic adsorbents into the peat-base inoculant†.

Time after inoculation (hrs)	Toxic seed Additives added to peat		Non-toxic seed	
	PVP	Charcoal	None	None
	-----Log no. rhizobia/seed-----			
0	5.8a*	5.6a	5.4a	5.6a
6	5.8a	5.6a	5.1b	5.6a
12	5.7ab	5.5a	4.8c	5.6a
24	5.6b	5.4a	4.6d	5.5a
48	5.0c	5.2b	4.2e	5.4b
72	4.2d	4.6c	3.3f	5.0c
96	3.7e	4.2d	2.3g	4.4d

*Means within a column followed by a common letter do not differ (P = 0.05) according to Duncan's Multiple Range Test.

†Seeds were incubated at 30°C and 100% relative humidity.

Breeding for genotypes producing non-toxic seed may be beneficial to ensure good survival and

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nodulation by the introduced strains. In the short-term, it may be useful to incorporate additives, such as charcoal, into the peat-base inoculants (10% of the peat weight) to neutralize the effect of the toxin, enhance survival and thus, increase the chances for nodulation and establishment.

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R. S. Kalmbacher¹

The probability of producers obtaining success in establishing legumes in grass-sod has not been proportional to the emphasis forage workers have placed on grass-legume associations. When all the important features of legumes are considered, one would expect the steps a producer follows to be well documented by research, and legumes would be relatively easy to consistently establish on a commercial basis.

Because of the expense involved in establishing legumes, it is important to follow certain steps to help protect the investment and assure a return. Differences in regions dictate that procedures for sod seeding legumes, which include site selection, pH and fertilization, seeding rate, date and variety recommendations, inoculation, seeding methods or machines, etc. must be decided at the local level. Local recommendations are designed to overcome limiting factors which are responsible for the inability of legume seeds to get beyond the seedling stage. Three major limiting factors within this first 30 to 45 days are light, soil moisture, insects and/or molluscs. It is my intention to discuss the effect that each of these factors has on seedling establishment. Hopefully, each forage worker will consider these effects and ways of overcoming them under their conditions.

Light

Legume seedlings are quite tolerant of low light (40% of incident) caused by shading from the grass canopy, and although low light is rarely responsible for outright death, seedlings probably never recover from low light intensity. The most visible effect of low light on seedlings is etiolation. Kalmbacher (unpublished data) found that hypocotyl length of *Aeschynomene americana* grown in 100% incident light was 9.8 mm, but increased to 11.5 mm at 75% incident; 16 mm at 14%; 22.5 mm at 27%; and 23.2 mm at 8% incident light. Etiolation is indicative of several other effects on seedlings that are less obvious such as: smaller roots, decrease in nodulation, decrease in relative growth rate and reduced leaf-stem ratio. In addition there may be some synergistic effects with soil moisture and herbivore pests, which will be considered later.

Smaller roots were found in 2-week old alfalfa seedlings grown in 2152 lux as compared with 4306 and 8611 lux, and shoot:root ratio varied

from 5.2, 3.6, and 2.6, respectively (Cooper 1966). When seedlings were 6-weeks old, root size was influenced less by reduced light. Shoot:root ratios were 1.5, 1.4, and 2.0 at these respective light intensities. Gist and Mott (1957) found that decreasing light reduced red clover and alfalfa root size. A 6-fold increase in light resulted in 2.5 and 5.0 fold increase in root weight of alfalfa and red clover, respectively. In a second study by Gist and Mott (1958) a 6-fold increase in light resulted in a 2.2 and 2.1-fold increase in weight of 15-day old alfalfa and red-clover seedling roots, respectively. Groya and Sheaffer (1981) demonstrated that shade always resulted in a reduction ($P < 0.05$) in root weight, regardless of soil moisture.

Decreased nodulation in alfalfa was found by Pritchett and Nelson (1951) when incident light was reduced from 2833 to 757 footcandles. This resulted in nodule number declining from 900 to 100 on 7, 70-day old plants. Decreasing light below 757 foot candles practically eliminated nodulation. Although reduced nodulation may not kill seedlings directly, it certainly would contribute to a reduced ability to survive and may add to slower growth rate.

A lower relative growth rate of alfalfa was found as light decreased (Cooper 1966). The decline was attributed to a decrease in the net assimilation rates of the legume seedlings. Growth of *aeschynomene* seedlings was reduced as incident light declined (Kalmbacher and Martin 1983). When grown in 100% incident light, seedlings weighed 45 g/50 seedlings; 41 g in 75%; 30 g in 45% incident light. These represent statistical, but probably not practical differences in legume size. Below 45% incident light, *aeschynomene* seedlings were greatly reduced in size. Like the effect of reduced nodulation, slower growth rates due to low light simply mean that legume seedlings are susceptible to other losses for a longer time.

Soil Water

The effect of soil water on sod-seeded legume development is less subtle, more direct, but more difficult to quantify than effects of low light. Insufficient water can inhibit germination, or there may be enough moisture for germination, but not enough to carry a plant through the seedling stage, in which case limiting water can kill seedlings directly.

Legume seedlings, although not as tolerant or competitive for moisture as established plants, are very tenacious. For example, 38% of the *aeschynomene* seedlings survived a 36-day period without additional water. Soil moisture was 12% at the start and 5.0% by weight (about 15 bars tension) at the end (Kalmbacher, unpublished).

Legume species have evolved under a wide range of conditions and have overcome many limitations. The following are some of these limitations and their effects on legumes.

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The legume seedling may begin life in one of the most hostile areas of the soil profile. Not only may light be limiting, but available soil water may be lowest in the upper few centimeters. Soil organic matter in an Ona fine sand at 0 to 3 cm depth was 8.7%, and at 0.3 and 15 bars tension contained 15% and 11% water, respectively (Kalmbacher, unpublished). Immediately below in the 4 to 6 cm soil depth, organic matter was 3.7%, and at 0.3 and 15 bars tension there was 8 and 6% water. At 13 and 15 cm depth there was 1.6% organic matter and 4 and 2% water at 0.3 and 15 bars tension.

Legume seedlings germinating under a low moisture regime have a difficult task of surviving, because not only do they germinate in a region where soil moisture may be held with greater tension, their roots begin in the upper soil where moisture extraction by the sod begins. A coastal bermudagrass sod in Georgia growing on a soil with a water content at 65% of field capacity in the upper 60 cm, lost 90% to 55% of that available water in the 0 to 15 cm depth, respectively (Doss et al. 1962).

Sod-canopy size may have an effect on available soil water through evapo-transpiration. On a soil which initially contained 65% of field capacity Doss et al. (1962) observed a 33% decrease in water evaporation after coastal bermudagrass or bahiagrass was cut to remove the canopy. On an Ona fine sand Kalmbacher (unpublished) found small differences in percent soil water when bahiagrass leaf area (canopy) increased. Bahiagrass with a LAI of 2.4 had an average of 4.9% water by weight as compared to 4.4% in bahiagrass with a LAI of 7.3. These percentages were an average of 5 stratifications in the upper 0-15 cm of soil which was very dry (about 15 bars tension). Under Florida's hot humid conditions evapo-transpiration may be similar on two largely different canopy sizes because high relative humidity could limit the process.

Reduced light and soil water together may have a synergistic effect, which results in seedling death. This would be a non-additive effect greater than the effect of moisture or light alone. Low light from a large grass canopy results in smaller legume roots (Cooper 1966, Gist and Mott 1957, Gist and Mott 1985, Groya and Sheaffer 1981), and if the large grass canopy brings about less available water through increased evapo-transpiration, then the competition from excessive cover could be compounded.

Insects and Molluscs

The canopy of a perennial grass provides an ideal habitat for insects, snails, and slugs which are often provided with a good food source growing in neat drill-rows. Seedling loss to these small herbivores is very rapid

and often complete, leaving the producer with the impression that germination never occurred.

When insecticide was used to control insects (primarily Orthoptera), ladino clover from a September seeding in North Carolina yielded 1,079 kg/ha vs 183 kg/ha when no insecticide was used (Rogers et al. 1983). In a second experiment demonstrating the merit of controlling insects, Rogers et al. (1983) had a 90% ladino clover stand that yielded 5,932 kg/ha when insecticide was used, but a 66% stand which produced 3,340 kg/ha without insecticide.

Snails and slugs have been a threat to successful legume establishment in North Carolina, Kentucky, Pennsylvania (personal communication) and perhaps in other states as well. In Florida the snail, *Polygyra cereolus*, at one snail/20 seedlings could result in 78% loss of white clover after 21 days (Kalmbacher et al. 1979). Field populations averaged about 10 snails/m², and such a population is a threat to any good stand numbering 150 to 200 legume seedlings/m².

A synergistic relationship between light and herbivore pests may exist. Control of a tall fescue canopy to allow more light to penetrate to September-seeded ladino clover in North Carolina resulted in the production of 2,560 kg/ha of clover, while use of insecticide with no canopy control resulted in 1,080 kg/ha (Rogers et al. 1983). When both insecticide and herbicide were used for canopy control, ladino clover yield was 5,570 kg/ha. When clovers were seeded in bahiagrass in Florida, (Kalmbacher et al. 1980) 1,310 kg/ha was produced with no snail control or canopy removal, but clover yield was 5,530 with canopy control (sod was disked) and 6,640 kg/ha with both canopy removal and control of snail (Paraquat and burn).

There are several reasons for this apparent synergistic effect. Eliminating the canopy increased light and resulted in legume seedlings which are not etiolated. Shorter hypocotyls may mean a seedling that is less vulnerable. Seedlings grow and develop more rapidly with more light and are susceptible to loss for a shorter time. Once a seedling develops the first trifoliate leaf it is less likely to be completely eliminated. Most importantly, more light may make the habitat less adequate for pests. A hot, dry, sunny soil surface for a slug is less desirable than a damp, cool habitat.

Practical Considerations

Light. In order to meet the light requirements of legumes the producer needs to determine when and if they need to control competition. The producer also needs to determine if canopy removal, cessation of canopy growth, or both are needed. A fall seeding in a subtropical grass may not need canopy control because cool weather will terminate growth, but there may be a large

canopy of senescent leaves that will produce the same results for developing legumes as a living canopy. A spring seeding on tall fescue will need canopy control and may also require removal of the canopy. These two distinct components of competition for light need to be recognized. The grazing animal can take care of both aspects; herbicides only result in termination of growth.

Water. Aside from irrigation, which may not be available, the only practical alternative at present is to identify the time which will maximize the probability of having adequate moisture. Robinson (1963) illustrates this by plotting the rainfall pattern for Savannah, Georgia against that of Stoneville, Mississippi. Ample precipitation was available through early September at Savannah, indicating that this would be a more likely time for successful legume establishment. The fall rains at Stoneville don't begin until mid November.

Perhaps in the future some synthetic acrylic polymers or super water absorbents seeded along with the legumes can hold enough water to tide seedlings over dry periods. This may be a promising area for some developmental research.

Pests. Producers need to determine the presence or absence of pests, and if they are present then insecticides, molluscicides or burning the sod before seeding may be prudent. Good canopy control and removal of cover may be a great benefit to minimize pest problems. Time of seeding may be very important. In North Carolina an October seeding has had less insect problems than a September seeding (Rogers et al. 1983). Fall seedings may be more risky than spring seedings, because the maturity and number of crickets, grasshoppers, etc. will be greater than a spring seeding.

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EVALUATION OF DESICCANTS FOR SODSEEDING CLOVERS

G. W. Evers¹

The benefits of growing cool season clovers are maximized when they are grown in mixtures with warm season perennial grasses in the Lower South. Animal performance improves because of the higher forage quality, nitrogen is added to the pasture system through symbiotic N₂-fixation, and the grazing season is extended. Climate and soils of the region limit the use of perennial legumes to the well drained, fertile soils. Cool season annual clovers have great potential for reducing livestock production costs on improved pastures throughout the South.

The greatest hazard for such a pasture system is the undependable establishment of the clover in the grass sod each fall. It makes little difference whether the clover is sodseeded or volunteers. The battle between a well established perennial grass and a young clover seedling for moisture, light and nutrients is always in favor of the grass. Management practices such as mowing, close grazing, and chemicals are used to improve clover seedling survival.

Paraquat and Glyphosate are the most widely used sod desiccants. Paraquat has quick activity but is not long lasting. The subtropical grasses begin to green up within 1 to 2 weeks after application. If the clover seedling does not emerge in 7 to 10 days, Paraquat has little benefit. Glyphosate is more effective and persistent. It is usually applied 3 to 7 days before seeding. However it can be phytotoxic to the perennial grass, especially dallisgrass and bahiagrass. Dalapon, which is sometimes used as a sod desiccant, is usually applied 3 weeks before planting and also has some phytotoxicity to dallisgrass and bahiagrass. However, it also lacks persistence on bermudagrasses with the grass frequently greening up 4 weeks after application.

A new class of herbicides is entering the market which are used for postemergence grass control in cotton and soybeans. Forage legumes appear to be quite tolerant of this herbicide group. A lower than recommended rate might be sufficient to desiccate a warm season perennial grass in the fall without affecting spring recovery. Since the chemicals can be applied pre- or post-emergence to the clover, they could also be used after volunteer clover stands emerge in the fall to improve clover establishment.

Several of these new chemicals were compared with the present herbicides used as desiccants on a dallisgrass sod (Table 1). Poast and CGA-82725 did a poor job of desiccating dallisgrass by planting time but did not retard spring recover. A Fusilade rate of .13 lb A.I./ac provided the correct balance of good desiccation and spring recovery of the dallisgrass. A similar study was initiated this past fall using several rates of the new compounds (Table 2). Paraquat provided excellent desiccation at planting time but was not persistent. Only 10% of the forage harvested on March 2 was ryegrass. Glyphosate, Dowco 453, and the .13 and .25 lb rate of Fusilade provided good desiccation and a high percentage of ryegrass. Desiccation rating on the day of planting was low for Dalapon but did improve as indicated by the 100% ryegrass at the first harvest. All other chemical treatments resulted in poor desiccation and low ryegrass percentages.

Poast, Fusilade, Dowco 453 and CGA-82725 were evaluated for any phytotoxicity to a pure stand of subterranean clover at 4 to 8 times the expected application rate (Table 3). The herbicides were applied when the clover seedlings were in the unifoliate to the first trifoliate leaf stage. There was no significant difference in seedling weight or first harvest yields.

Of the herbicides assessed, Fusilade and Dowco 453 have the greatest potential for use as grass sod desiccants in aiding clover establishment. At this time, however, none of these chemicals have been evaluated on grass sods overseeded to clovers.

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Table 1. Evaluation of herbicides on dallisgrass for sodseeding 1982-83.

Herbicide	Rate (A.I.)	Application before seeding	Desiccation ¹ rating	Recovery ² rating
	1b/ac		Oct. 20	Aug. 1
Control	--	--	1.00	5.00
Paraquat	.5	1 day	4.75	5.00
Glyphosate	.5	1 wk	3.75	.75
Dalapon	3.75	3 wk	4.25	1.00
CGA-82725	.13	3 wk	1.00	4.75
Poast + C.O.	.13	1 wk	1.75	4.75
Fusilade + C.O.	.06	3 wk	2.25	4.50
Fusilade + C.O.	.13	3 wk	4.00	4.00
Fusilade + C.O.	.25	3 wk	3.75	1.75

¹Desiccation rating 1-no desiccation, 5-complete desiccation.²Dallis recovery rating 0-no dallis, 1-<10% dallis stand, 2-25% dallis stand, 3-50% dallis stand, 4-75% dallis stand, 5-100% dallis stand.

Table 2. Evaluation of herbicides on dallisgrass for sodseeding 1983-84.

Chemical	Rate (A.I.)	Application before seeding	Desiccation ¹ rating	Ryegrass ²	
	1b/ac			%	1b DM/ac
Control	--	--	0.00	6	30
Paraquat	.5	1 day	4.00	10	28
Glyphosate	.5	1 wk	5.00	100	527
Dalapon	3.75	3 wk	1.75	100	541
Fusilade	.06 + C.O.	3 wk	2.00	14	110
Fusilade	.13 + C.O.	3 wk	4.50	72	437
Fusilade	.25 + C.O.	3 wk	5.00	100	607
CGA-82725	.13 + C.O.	3 wk	2.75	14	54
CGA-82725	.25 + C.O.	3 wk	2.75	22	125
Dowco 453	.06 + C.O.	3 wk	5.00	100	453
Dowco 453	.13 + C.O.	3 wk	5.00	100	364
Poast	.13 + C.O.	1 wk	1.00	11	44
Poast	.25 + C.O.	1 wk	2.25	18	168

¹Desiccation rating November 2, 1983, 1-no desiccation, 5-complete desiccation.²Data collected March 2, 1984.

Table 3. Effect of post emergence herbicides on subterranean clover seedling weight and first harvest yields.

Herbicide	Rate	Seedling weight ²	First harvest
	1b A.I./ac	mg	1b D.M./ac
Control	--	243a ³	2111a
Poast	.5	240a	1843a
Poast	1.0	208a	2136a
Fusilade	.5	208a	2211a
Fusilade	1.0	233a	1943a
Dowco 453	.5	275a	1676a
Dowco 453	1.0	223a	1843a
CGA-82725	.5	245a	2136a

¹Herbicides applied when clover was in unifoliate to first trifoliate stage.²Seedling weights determined 7 weeks after planting.³Values within a column followed by the same letter are not significantly different at .05 level, Duncan's Multiple Range Test.

NO-TILL ESTABLISHMENT OF CLOVERS IN DALLISGRASS AND BAHIA GRASS

R. W. Taylor¹, J. L. Griffin¹, and G. A. Meche¹

In the Gulf Coast states the predominant pasture species include bermudagrass [*Cynodon dactylon* (L.) Pers.], bahiagrass (*Paspalum notatum* Flugge.), and dallisgrass (*P. dilatatum* Poir). At present, only limited acreage has been improved by the introduction of legumes. Volunteer legume stands constitute the majority of the legume-grass acreage. Grassland pasture renovation by seeding legumes using no-till techniques offers an alternative for pasture renovation. Sod-seeding of legumes in grass sods should increase sward productivity and quality. Addition of legumes to grass sod may also extend the grazing season and supply nitrogen for associated grass species. A series of experiments were conducted in order to evaluate legume species for establishment and productivity when sod-seeded into dallisgrass and bahiagrass swards and to assess the effectiveness of cultural practices on legume establishment.

Eight legume species were evaluated for potential suitability for no-tillage renovation of dallisgrass and bahiagrass sods. Legume species were as follows: Alfalfa (*Medicago sativa* L. cv. Cimarron), red clover (*Trifolium pratense* L. cv. Redland II), white clover (*T. repens* L. cv. Regal), persian clover (*T. resupinatum* L.), subterranean (sub) clover (*T. subterraneum* L. cv. Mt Barker), blue lupine (*Lupinus angustifolius* L. cv. Tifblue-78), white lupine (*L. albus* L. cv. Tifwhite-78), and arrowleaf clover (*T. vesiculosum* Savi cv. Yuchi). Sub clover from the 1981 fall seeding was allowed to naturally reseed and for the 1982-1983 production season was included as an additional treatment referred to as reseeding sub clover. A non-seeded grass control was also included. Randomized, complete block designs with four replications were used. Legumes were seeded during the period 30 Oct. to 6 Nov. in 1981 and 22 Oct. to 29 Oct. in 1982.

The effects of cultural practices on no-till legume establishment in dallisgrass and bahiagrass sod were also evaluated. A split plot design with subplots applied as strips across main plots (split block) with three replications was used. Main plots consisted of eight cultural practices: No legume seeded (control); mowed and drill seeded; mowed and broadcast seeded; mowed, broadcast seeded, and disked; paraquat applied (0.56 kg ai/ha) and drill seeded; paraquat applied (0.56 kg ai/ha), residue burned, and drill seeded; glyphosate (Roundup) applied 2.24 kg ai/ha), and drill

seeded; and sethoxydim (Poast) applied (0.45 kg ai/ha) and drill seeded. Subplots were legume species: Alfalfa (cultivar-Baron) (1981 only), sub clover drill seeded in the fall with cultural treatments fall-applied (1981 and 1982), Mt Barker clover allowed to reseed naturally from previous year's treatments (no cultural treatments applied in Oct. 1982) (1982 only) replaced alfalfa, and Redland II red clover drill seeded with cultural treatments fall-applied (1981 and 1982). In 1982 Redland II red clover seed had been pelleted with *Rhizobia* and lime; so therefore, the effective seeding rate was found to be reduced by 30%. All seed were inoculated with the appropriate rhizobia bacteria prior to seeding. Sub clover allowed to reestablish from the previous season's seed production will hereafter be referred to as reseeding sub and sub clover and red clover seeded each year will be called fall-seeded sub or red clover.

The experimental site was a Crowley silt loam (Typic Albaqualf) soil. Harvesting was timed according to species and forage availability and January to October the year following fall seeding. At each harvest when seeded species were present, botanical estimates of percent biomass for seeded species and grass plus weeds were obtained by visual observations and by hand separation of a sample harvested for botanical composition. Hand separations of material from 1 to 2 ft² were made and used to determine dry matter yield of seeded species and the grass plus weed components.

Averaged over two years, pure legume yield (based on botanical separations) of the eight legume species seeded in dallisgrass sod was ranked as follows: Red clover = arrowleaf clover > reseeding sub clover ≥ fall seeded sub clover ≥ white clover = persian clover > white lupine = blue lupine = alfalfa. Relative rankings of legumes sod seeded in bahiagrass were similar except that sub clover yields were generally greater than white clover, persian clover, and blue and white lupine. In bahiagrass alfalfa yield was less than that of all other legumes. Unusually low yields from alfalfa were possible due to the low soil pH whereas blue lupine yields were reduced because of persistent grazing by rabbits. White lupine yields were influenced by poor stands in part due to the large seed size.

Total season production (legume plus grass) averaged across years for legumes seeded no-till in dallisgrass sod were ranked as follows: Arrowleaf clover = red clover > reseeding sub clover = persian clover ≥ alfalfa = blue lupine = white clover = white lupine = fall-seeded sub clover > grass control. In bahiagrass sod arrowleaf and red clover were again superior to the other legumes which were all superior to the grass control.

In summary, the most promising species for no-till sod seeding in grass sods were red, arrowleaf, and sub clover. No-till legume establishment showed potential for increasing

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the productivity of native grassland pasture. Sod seeding of legumes was generally more productive in dallisgrass than bahiagrass. Sub clover showed promise as a reseeding legume for pasture use.

Cultural practices had a significant influence on no-till establishment of fall-seeded sub and red clover and reseeding sub clover in dallisgrass and bahiagrass sods. In 1982 alfalfa establishment was variable for both sods and all cultural treatments; therefore, alfalfa yields will not be discussed. Legume production in 1982 was greatest for red clover although total season production (legume plus grass) was similar between fall-seeded red and sub clover. Total season production in 1982 for fall-seeded red clover averaged over grass sods was ranked as follows among cultural treatments: Drill-paraquat-burn \geq drill-Poast = drill-Roundup = drill-no herbicide = drill-paraquat > broadcast = broadcast-disk grass control. Total season production in 1982 for fall-seeded sub clover averaged over grass sods was ranked as follows among cultural treatments: Drill-Poast = drill-paraquat-burn = drill-paraquat = drill-no herbicide = drill-Roundup > broadcast = broadcast, disk \geq grass control.

In 1983 on dallisgrass sod, total production from all species for all cultural treatments was greater than that for the control treatment. Total yield of fall-seeded red clover in dallisgrass was ranked as follows: Drill-no herbicide = drill-paraquat \geq drill-paraquat-burn = broadcast, disk = drill-Poast = broadcast = drill Roundup. For fall-seeded sub clover total season production in dallisgrass was ranked as follows: Drill-Poast = drill-paraquat-burn = drill-paraquat = drill-no herbicide > drill-Roundup \geq broadcast = broadcast-disk. Total forage yield was lower for broadcast followed by light disking than for drilling without herbicide or with Poast or Roundup for reseeding sub clover. Averaged over legume species, total season production from cultural practices was ranked as follows: Drill-no herbicide = drill-paraquat = drill-Poast = drill-paraquat-burn \geq drill-Roundup \geq broadcast = broadcast-disk > control.

On bahiagrass sod in 1983, fall-seeded red clover total season yields (legume plus grass) were not significantly different among cultural treatments. For fall-seeded sub clover drill seeded with paraquat, total season yield was greater than that for other cultural treatments except drill-paraquat-burn. Total yield of fall-seeded sub clover broadcast followed by light disking was not significantly different from the grass control but significantly less than that for other cultural treatments. Reseeding sub clover yield was greatest for the cultural treatments drill-no herbicide, drill-Poast, and drill-paraquat burn. Total yield for reseeding sub clover broadcast-disk and drill-Roundup was lowest. Averaged over legume

species, yield of the broadcast-disk treatment was significantly lower than for drill-no herbicide, drill-paraquat-burn, and drill-Poast.

In summary no-till legume establishment in dallisgrass and bahiagrass sod significantly increased total season yield above the grass control. Generally, drilled legumes produced significantly greater legume and total season forage yield than broadcast seeded legumes. No-till legume seeding appeared to be a viable alternative to broadcast overseeding of grass pastures.

PARTICLE SIZE AND FORAGE EVALUATION

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INTRODUCTION

Forages are developed by 1) selection for desired agronomic characteristics and 2) selection for improved utilization and performance by the animal. Improving agronomic characteristics involves selection for persistence, yield and quality (in vitro digestibility and crude protein content) and animal evaluation can involve palatability, preference, intake and weight gain. Since the animal is the ultimate forage consumer, more emphasis needs to be placed on animal evaluation of the forage early in the selection process. The physical processing of the forage and resulting particle size reduction accomplished by the animal has received only limited attention in evaluating the forage-animal interface.

Animals consuming hay or grazing, breakdown the forage to smaller particle sizes to allow greater microbial accessibility and fermentation thus increasing the probability for exiting the reticulo-rumen. Therefore, ease of size reduction may be related to forage intake, digestibility and utilization by the animal.

MASTICATION EFFECTS

Particle size reduction begins with prehension and initial mastication of the forage. Pond et al. (1984), using esophageal cannulated cattle, reported the wide range of particle sizes derived solely by initial mastication. These results show when grazing Coastal bermudagrass more than 50% of the particles (by weight) passed through a 1 mm screen. Light microscopy identified plant parts and tissues associated with each particle size and also the sites of physical fragmentation and rupture. In addition to particle size reduction, mastication also resulted in crushing and crimping of intact particles to further increase their surface area and microbial enzyme accessibility. Without such physical disruption the microflora of the reticulo-rumen would be unable to penetrate the barriers formed by the cuticle and vascular tissue. Many small particles were

highly lignified with little potentially digestible tissues present. Plants are not homogeneous, therefore, particle size should be evaluated by plant part and identified as to anatomical origin. Tissue exposure and composition of similar size particles can be dramatically different.

Extreme differences due to mastication existed between forages. Ryegrass, although having larger particles, differed from Coastal bermudagrass in cuticle attachment and ease of tissue exposure. The cuticle of Ryegrass appeared to be more fragile and less firmly associated with the leaf blades. After mastication, large sheets of cuticle with attached and exposed mesophyll and epidermal tissue were present. The cuticle of Coastal bermudagrass offered a more protective barrier.

PARTICLE SIZE AND DIGESTION

Malooji et al. (1982) and Pond et al. (1982) utilized intact particles of Coastal bermudagrass hay obtained after initial mastication and determined in vitro rate, lag time and extent of neutral detergent fiber (NDF) digestion for seven different particle sizes.

The rate and extent of NDF digestion was highest with the small particles (20 to 100 μ m) and decreased as particle size increased. The lag time ranged from 0 h for small particles to 6 h for particles greater than 1700 μ m. Large particles appear to require a longer time before digestion begins and have a slower rate of digestion; therefore they are digested to a lesser extent.

Caution is needed in conducting and interpreting digestion studies involving different particle sizes. 1) To determine the true relationship between particle size and rumen digestion, particles should be derived from mastication. Grinding or mechanical cutting of forages will not mimic the mastication process of the animal (Pond et al. 1984). 2) Total extraction of the cell solubles of intact large particles is not possible, therefore overestimation of fiber content is probable. After digestion large particles need to be reduced in size to allow complete extraction of cell solubles. 3) The porosity of gooch crucibles (coarse porosity) is 40-60 μ m. Filtering samples containing fine particles could underestimate material retained by the crucible and therefore underestimate NDF values of small particles. Alternative filtering procedures or use of fine porosity gooch crucibles is required for these small particles.

PARTICLES IN THE RUMEN

Baker and Harriss (1947) reported on the microscopic observation of digesta contents of ruminants and Evans et al. (1973) reported some of the physical characteristics of the digesta in the reticulo-rumen of cattle. Generally,

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across all forages, larger particles are associated with the dorsal sac of the rumen and mean particle size decreases as the rumen strata is sampled ventrally. Digesta density is lowest in the dorsal sac and highest in the ventral sac and reticulum. Pond (1982) determined for Coastal bermudagrass that within a particle size there was no difference between particles obtained in the dorsal vs. ventral sac when viewed with the light microscope. Several scientists have suggested that particles must be reduced in size to 1000 μ m to pass from the rumen. However, particles twice that size are often found in the feces. Although particle size is important in determining exit from the rumen, it is not the complete answer. Other factors as density of material, chemical composition, structural anatomy, amount of air entrapment and rate of hydration are probably involved.

FRAGMENTATION OF FORAGES

Little is known about how intact plants are fragmented to smaller particles. The physical breakdown and fragmentation of Coastal bermudagrass was described by Pond (1982). The progressive pattern to smaller fragments is presented in figure 1.

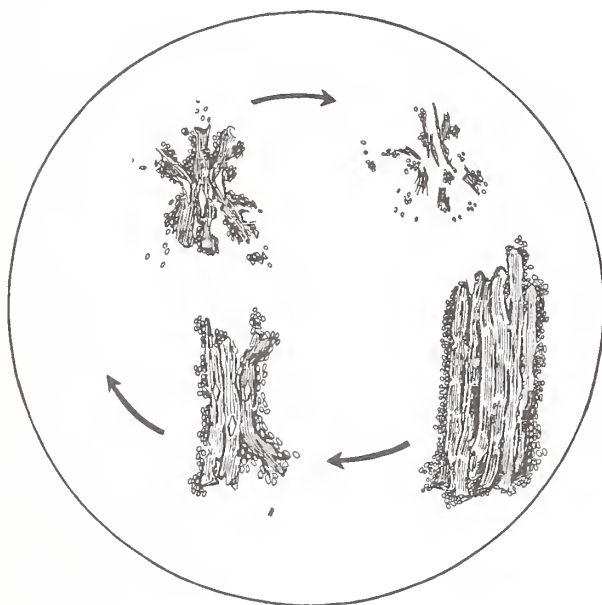


Figure 1. The fragmentation of Coastal bermudagrass.

The rectangular dimensions of successively smaller fragments appear to be determined by the number of associated vascular bundles (determining width) and vascular bundle length (determining length). Fracturing occurred along bundles and bundles were severed at the ends. Smaller particles appeared to be derived by further reduction in length and number of associated bundles. Splits occurred between bundles increasing tissue exposure and

the smallest fragments were derived by further disruption of the remaining plant tissue.

Similar fragmentation patterns may exist for most grasses but legumes would have different fragmentation patterns. Troelsen and Cambell (1968) described alfalfa hay digesta to be comprised of short broad particles whereas grass hay was longer, thinner and more fiber-like. Differences in the anatomical structure of plants may explain differences in fragmentation characteristics. Most grass leaves contain parallel vascular bundles which seem to determine where fragmentation occurs whereas legumes have a branched network of vascular tissue which might expectedly yield rectangular fragments of near equal length and width. More work is needed in the area of forage fragmentation to determine differences due to plant type, maturity and species.

FUTURE EVALUATION

Several factors in combination may be appropriate for future forage development and evaluation. Relating the physical properties of the forage (Seoane et al., 1982), the effects of chewing on the physical breakdown (Reid et al., 1979) and tying in the effects of chewing on particle size reduction and subsequent fermentation (Ulyatt et al., 1982) should all be very important in future forage-animal evaluation work. This will require the combined effort of plant breeders, agronomists, plant physiologists, animal scientists and when surgically modified animals are required, veterinarians.

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BEEF-FORAGE SYSTEMS FROM CONCEPTION TO SLAUGHTER

D. G. St. Louis¹

INTRODUCTION

Beef-forage systems research was conducted at the Edisto Experiment Station near Blacksville, SC. The study was divided into cow-calf systems, backgrounding or stocker steer systems and finishing steer systems. The systems were designed for intensive land use and maximum recovery of available forage and to optimize quantity and quality of grazing. The level of nutrition provided by the forage systems was designed to meet the requirements of the various classes of animals.

COW-CALF SYSTEMS

The cow-calf systems were closed in that the number of animals and land area remained constant throughout the year. Data on year-round forage production and utilization were collected. Any cow without a calf at side at the beginning of the breeding season on April 1 was removed and replaced with a cow with calf at side. All open cows were culled at weaning in early September and were replaced with pregnant cows. Cows were bred in a 60-day breeding season which resulted in a January and February calving season. Calves were weaned near September 1 each year at an average age of 7 months. Each cow-calf system was composed of 9.45 ha and 24 cows. Cow-Calf System 1 (CC1) was based upon sorghum silage and Coastal bermudagrass pasture. One-third of the land area produced forage sorghum for silage in the summer and rye pasture in the winter. The other two-thirds of the land area produced Coastal bermudagrass pasture in the summer and was overseeded with rye and Yuchi arrowleaf clover in the fall for spring grazing. Cows were supplemented with sorghum silage in drylot when winter grazing was inadequate. Calving was in drylot so that cow-calf pairs were turned to rye pasture as calves were born. Cows were kept on Coastal bermudagrass pasture in the fall until grazing was inadequate and the stubble would not interfere with sod seeding of rye and arrowleaf clover. Sod-seeding planting dates varied between October 15 and December 1 with equal success.

Cow-Calf System 2 (CC2) was similar to traditional cattle operations based upon Coastal bermudagrass hay and pasture. This land area was divided into two thirds so that pastures and hay fields could be rotated. Rye and arrowleaf clover were planted on two-thirds of the land area as in CC1. Hay

feeding and calving were on the field to be used for hay production the following summer. This rotation proved effective in weed control.

Both cow-calf systems produced adequate stored feed to meet the winter feeding requirements. Winter feed production was deficient in one year and in excess in two years. Weaning weights of calves were not significantly different between the systems with a combined mean of 196 kg. Cows in CC1 carried slightly more body condition throughout the year than cows in CC2 primarily due to rye grazing in the fall. Limit feeding of sorghum silage was necessary in CC1 to prevent cows from becoming too fat prior to calving. The primary differences between the systems were economic. Although economic analyses are incomplete, indications are that the silage based system should be about double the size of CC1 to provide economy of scale for silage production equipment in order to be competitive. CC1 seems to be suitable for an earlier calving period but supplemental grain might be required in the breeding season.

Phosphorus and potassium fertilizer were applied according to soil test to insure that they did not limit forage production. Rye on prepared land was topdressed in the fall with 60 kg/ha of nitrogen. In February or March 60 kg/ha of N was again applied to all winter annual pastures for spring grazing. Nitrogen was applied to Coastal bermudagrass and millet pastures at the rate of 200 kg/ha of N in two applications, to sorghum at 100 kg/ha in one application and to Coastal bermudagrass hay fields at 200 kg/ha in April and again after each cutting taken before September 1.

BACKGROUNDING-FINISHING SYSTEMS

Backgrounding and finishing systems for steers were studied in component part of a year-round forage program. Phase 1 was from weaning until winter grazing, phase 2 was winter grazing, phase 3 was from winter to summer grazing and phase 4 was summer grazing which terminated when steers went to slaughter. Some of the steers were produced outside of the cow-calf systems to maintain adequate numbers. Sorghum silage and limited grain were fed when grazing was not available. Rye pasture was grown in rotation with forage sorghum or millet pasture and fertilizer at the rates described for cow-calf systems. Phases 2 and 4 which are the grazing components are summarized here.

Backgrounding

Backgrounding treatments were rye pasture at a stocking rate of 4.9 hd/ha (BG1), rye pasture and Coastal bermudagrass stubble at a stocking rate of 2.5 hd/ha (BG2), rye pasture and stubble at a stocking rate of 4.9 hd/ha (BG3) and drylot feeding of sorghum silage and concentrates (BG4). BG2 and BG3 each had 1/2 of the land areas devoted to rye pasture and Coastal bermudagrass stubble. If the area in stubble

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were ignored, BG1 and BG2 had equal stocking rates of rye pasture while BG3 had double the stocking rate of the first two treatments. BG4 was designed as a base for comparison with the pasture treatments. Wren's Abruzzi rye was planted between September 27 and October 14. Grazing began between November 11 and December 7. Much of the time difference between planting and grazing was due to the annual differences in temperature and rainfall but there was a clear advantage to planting early. Coastal bermudagrass was grazed until September 1 so that whatever remained, plus regrowth, determined the amount of stubble for this study. Due to frost, little green grass was available when grazing began.

When grazing was not adequate, sorghum silage was fed ad libitum plus .45 kg of a 44% protein supplement. The amount of corn fed varied between .30 and 2.27 kg/hd/da depending upon the amount of grazing judged to be available. It is felt that the greater amount of corn should have been maintained whenever it was necessary to feed sorghum silage. All steers received monensin at 200 mg/hd/da throughout the study. The drylot ration for BG4 was similar to that used to supplement inadequate pasture and was designed to produce similar gains. A lesser amount of corn (1.82 kg/hd/da) was fed to steers in BG4 of the first trial, but 2.27 kg/hd/da was fed thereafter.

The total grazing days ranged between 104 and 149 for four trials. It was impractical to extend grazing beyond April 1 because millet and sorghum had to be planted and rye pasture quality had decreased rapidly due to maturation. If Coastal stubble had been beneficial, it would have increased the number of grazing days or decreased the amount of supplement required. The difference between 61 and 62 days of BG1 and BG2, respectively, was not significant for the grazing days that no supplement was required. BG3 with double the stocking rate of BG2 produced only one-half the number of grazing days of BG2. The greatest ADG was .90 kg for BG1 but this was not significantly different from BG2 with .87 kg. Other differences were significant with .60 kg for BG3 and .81 kg for the drylot treatment. Errors in judgment were made as to when steers in BG3 needed supplementation, otherwise the gains in BG3 should have equalled or exceeded BG4. The initial weight of steers were 194, 187, 239 and 267 kg while the final weights were 297, 352, 310, 383 kg for the first through fourth trials, respectively. The gains varied by years primarily due to the effects of weather conditions on forage production.

Table 1. Supplementation of backgrounding steers¹

Description	Units	Treatments			
		BG1	BG2	BG3	BG4
Total supplement ²	kg/hd	447	448	579	1114
	kg/hd/da	5.38	5.81	5.73	8.63
Percent of BG4	%	62.3	67.3	66.4	100.0
Days supplemented	da	83	84	101	129
Equivalent days ³	da	52	57	67	129
Average daily gain	kg	.96	.95	.68	.78
Gain	kg/hd	124	123	87	100
Supplement ⁴	kg/hd	40	44	52	100
Pasture ⁵	kg/hd	84	79	35	0
Gain	kg/hd	247	246	349	-
Supplement	kg/hd	81	88	209	-
Pasture	kg/hd	166	158	140	-
Efficiency ²	supp/gain	3.62	3.97	6.65	11.11

¹Data from 3 of 4 trials as drylot treatment not included in the first backgrounding trial.

²Dry matter supplementation included all concentrates and sorghum silage.

³Percent of BG4 as percent of days supplemented.

⁴ADG of .78 kg times equivalent days.

⁵By difference from total gain.

Table 1 compares pasture treatments with BG4. Equivalent days were calculated by taking the percent of BG4 as a percent of days supplemented. Assuming that equivalent days more closely represented the days that supplement would be fed if steers had been taken off pasture when supplemented, that supplementation for the equivalent days would result in the same ADG as BG4, amount of gain due to supplement. Gain due to pasture was the difference between total gain and gain due to supplement. Differences between BG1 and BG2 in gain due to pasture did not appear to be different. In BG3 gain due to supplement was greater than gain due to pasture. Ignoring the area in stubble, the gains per hectare due to pasture did not differ greatly. If the heavier stocking rate of BG3 were beneficial, then gain per hectare due to pasture should exceed the other two pasture treatments. This was not true because overgrazing probably hindered regrowth. The supplement-to-gain ratio which included sorghum silage showed the trends that should be expected. There was no advantage of Coastal stubble on feed efficiency at the lighter stocking rates.

Finishing

The same steers used in the backgrounding systems were reassigned to finishing systems. The finishing treatments were millet pasture at 4.9 hd/ha (ML1), millet pasture at 9.9 hd/ha

(ML1), millet pasture at 9.9 hd/ha (ML2) Coastal bermudagrass pasture at 4.9 hd/ha (CB1) and Coastal bermudagrass pasture at 9.9 hd/ha (CB2). Tifleaf or Gahi-3 hybrid pearl millet was planted between April 19 and May 10. Summer grazing began between May 31 and June 16. Again, planting early optimized the quantity and quality of millet grazing. Grazing of millet began when it was approximately 15 cm tall. Coastal bermudagrass grazing began between May 31 and June 8 when it was approximately 10 cm tall. In the second trial, dry weather delayed grazing of millet by 8 days compared to Coastal bermudagrass. Coastal bermudagrass was an old established stand mixed with bahiagrass. Ground shelled corn and a mineral-moensin premix was fed at 1% of body weight and adjusted biweekly when steers were weighed. Steers were on pasture from 94 to 99 days. Initial weights showed a yearly increase from 321 kg in the first year to 372 kg in the third year. The mean ADG of 1.09 kg in the first year was not significantly different from 1.08 kg in the third year. The lower ADG of .84 kg in the second year was significantly different from the first and third years primarily due to dry weather and reduced pasture growth. Final weights were 423, 449, and 479 kg for the first through third trials, respectively.

Steers grazing millet had an ADG of 1.08 and 1.10 kg for ML1 and ML2, respectively, while steers grazing Coastal bermudagrass had an ADG of .95 and .91 kg for CB1 and CB2, respectively. Utley et al. (1976) found similar differences between millet and Coastal bermudagrass with put-and-take grazing and no grain. The differences in ADG between stocking rates of 4.9 and 9.9 hd/ha were not significant for either type of pasture suggesting that stocking rates were not great enough for reduced forage availability to affect animal gains. This was supported by the need to clip pastures to maintain quality grazing. Woods and Suman (1966) observed a mean stocking rate of 10.9 hd/ha on Coastal bermudagrass with put-and-take grazing and supplementation with corn at 1% of body weight. Limited forage availability in the second trial resulted in a significant treatment by year interaction in ADG. In this year of dry weather millet did not significantly outperform Coastal bermudagrass. Also, Coastal bermudagrass at the lower stocking rate of CB1 showed significantly greater ADG than CB2. Gains per hectare primarily reflect differences in stocking rates with 514, 1055, 451, and 870 kg/ha for ML1, ML2, CB1 and CB2, respectively. The difference between ML1 and CB1 was not significant while the difference between ML2 and CB2 was significant.

Steers in the first trial had light initial weights and did not reach satisfactory final weights or grade at slaughter. Hot carcass weights of less than 225 kg were discounted \$.09/kg at the packing plant. In the first

trial, 50% of carcasses fell below this weight while 4.4% of the carcasses were too light in subsequent trials. Quality grades across trials primarily reflected the carcass weights. In the first trial, 47% of the carcasses graded Standard resulting in a \$.03/kg reduction in price below the Good grade. In subsequent trials, carcasses graded an average of low Choice. Year effects were significant for all carcass parameters measured except lean color and fat firmness. In the second trial, 15% of the carcasses were discounted \$.43/kg for having yellow fat. Treatment effects were not significant for any carcass parameters except yield grade. This indicated that some of the weight gain differences between millet and Coastal bermudagrass pastures were due to body fat accumulation.

Conclusions

Gains on pasture may have been close to maximum but profits for finishing steers were not competitive with drylot finishing. In addition, a drylot feeding period may be necessary to overcome packer and consumer objections to yellow fat. With the variation in weaning weights, calves should be sorted into weight groups and various alternatives for backgrounding should be used. It is then possible to begin drylot finishing of steers weighing at least 320 kg at various times during the year. Some heavy steers should begin drylot feeding at weaning and others should begin after the winter grazing period. Lightweight steers should be backgrounded through both the winter and summer grazing periods before finishing begins.

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W. R. Windham¹

INTRODUCTION

The quality of a forage is determined by the chemical composition and physical characteristics. Therefore, laboratory analyses are essentially aimed at obtaining analytical data that indicate forage quality and predict the rate and extent of biological degradation. The analytical problem becomes that of determining the factors in the forage which limit degradation; hence, there is emphasis on lignins and other components of the fibrous cell wall that provide resistance to digestion. Several schemes of analysis have been developed to determine chemical composition of forages. Of these, the systems of fibrous feed analysis devised by P. J. Van Soest and coworkers have become the primary standards for chemical evaluation of forages. This system of detergent methods fractionates the forage fiber relative to its nutritive availability and recognizes the distinction between cell walls and cell contents. Neutral detergent fiber (NDF) appears to separate the soluble forage constituents (cellular contents) from those that depend on microbial fermentation (cell walls), (Goering and Van Soest, 1970). The NDF fraction is considered to be essentially hemicellulose, lignocellulose and insoluble ash. Acid detergent fiber (ADF) divides forage components into fractions soluble and insoluble in 1 N acid. The acid-soluble fraction is primarily the hemicellulose and cell wall proteins, while the acid-insoluble fraction is lignocellulose and silica. Therefore, the difference between neutral- and acid-detergent fiber residues was proposed as an estimate of hemicellulose.

The detergent system has also been extended to the analyses of in vitro residues, abomasum particulate and fecal residues. Based on the analysis of the indigestible residues, apparent digestibility coefficients have been reported for NDF, ADF, hemicellulose and cellulose. In addition, the lignin content of the ADF residue has frequently been used as a marker to determine the digestibility of forage dry matter. However, recent data indicates possible degradation and solubilization of lignin in the digestive tract resulting in incomplete lignin recovery and underestimation of forage digestibility. The purpose of this paper is to review systems of analysis for fiber specifically hemicellulose and to discuss the effect of analysis type on estimation of cell wall constituents in in vitro residues and fecal residues.

Hemicelluloses are cell wall structural polysaccharides which comprise a significant portion of the dry matter of tropical and temperate forage with lesser amounts present in legumes and represent a major potential source of energy to ruminants. Often forage components are classified according to the analytical procedures used to separate given polysaccharides. These procedures generally fall into two categories: a) those involving hydrolysis of the cell wall, and b) those involving extraction with various solvents. In addition, the difference between neutral- and acid-detergent fiber residues was proposed as an estimate of hemicellulose (i.e., conventional analysis). However, interferences in the estimate of hemicellulose by difference have been reported (Van Soest and Robertson, 1979) due to preferential solubility and/or precipitation of various forage fractions in neutral detergent (ND) and acid detergent (AD) reagents. Bailey and Ulyatt (1970) reported that at least one-half of the pectic substances are not removed and much of the hemicellulose is not extracted with 1 h AD treatment which would result in a higher estimate of ADF and a lower estimate of hemicellulose. Van Soest and Robertson (1979) reported that pre-extraction with ND followed by AD (i.e., sequential analysis) would allow a more accurate estimate of hemicellulose.

Direct estimate of hemicellulose from hydrolysis of plant cell walls (i.e., NDF) with trifluoroacetic acid (TFA) has been reported by numerous researchers (Collings and Yokoyama, 1979; Barton et al., 1982; Widham et al., 1983; Widham and Amos, 1984). Upon hydrolysis of NDF by TFA, hemicellulose was assumed to be the acid soluble portion of NDF (HC-TFA) and cellulose plus lignin (ADF-TFA) the acid insoluble portion of NDF. Windham et al., (1983) and Windham and Amos (1984) reported that HC-TFA and ADF-TFA values were significantly different from the ADF and hemicellulose obtained by conventional detergent analysis (Table 1). However, sequential analysis resulted in estimates of ADF and hemicellulose that resemble estimates obtained by hydrolysis of NDF with TFA. The effect of analyses type on the determination of cell wall constituents results in an analysis type by forage interaction ($P < .03$). This interaction, or lack of consistent extraction across forages by methods illustrates the difficulty of designing a single system of analysis for all conditions. Therefore, the choice of analytical methods may drastically affect the interpretation of cell wall fiber data.

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Table 1. Effect of analyses type on estimation of forage cell wall constituents

Forage	Analysis	Hemicellulose	Acid Detergent Fiber	Cellulose	Lignin
------(%)-----					
Kentucky-31 tall fescue	Conventional	25.8a ^{1/}	27.4a	24.4a	3.0a
	Sequential	26.0a	26.6a	23.7a	2.9a
	TFA hydrolysis	26.9a	26.3a	23.4a	2.9a
Coastal bermudagrass hay	Conventional	33.6a	33.6a	29.9a	3.7a
	Sequential	37.8b	31.8b	28.8b	3.0b
	TFA hydrolysis	38.5b	31.8b	27.9b	3.1b
Alfalfa hay	Conventional	12.1a	36.6a	27.9a	8.7a
	Sequential	15.2b	32.1b	24.0b	8.0b
	TFA hydrolysis	17.8c	30.2c	23.0c	7.2c

^{1/} a,b,c Means within columns within forage with unlike superscripts differ (P .05).
Adapted from Windham et al., 1983 and Windham and Amos, 1984.

Table 2. Effect of analyses type on estimation of forage cell wall constituents in fecal particulate digesta.

Forage	Analysis	Hemicellulose	Acid Detergent Fiber	Cellulose	Lignin
------(%)-----					
Coastal bermudagrass hay	Conventional	25.2a ^{1/}	35.7a	24.5a	11.2aa
	Sequential	29.8b	30.8b	23.9a	7.0b
	TFA hydrolysis	30.9b	29.0b	20.8a	8.2b
Alfalfa hay	Conventional	12.6a	49.3a	31.7a	17.6a
	Sequential	19.1b	43.0b	29.6a	13.4b
	TFA hydrolysis	24.8c	36.7c	24.8b	11.9c

^{1/} a,b,c Means within columns within forage with unlike superscripts differ (P .05).
Adapted from Windham and Amos, (1984).

EFFECT OF ANALYSES TYPE ON FERMENTATION RESIDUES

The effect of analyses type on the estimation of forage cell wall constituents in fecal particulate digesta of steers fed Coastal bermudagrass and Alfalfa hay are shown in Table 2. A type of analysis by forage interaction was found for hemicellulose and ADF. This interaction was also observed in the analyses of the forages (Table 1), however, the magnitude of the increase ($P < .05$) in hemicellulose and decrease ($P < .05$) in ADF are greater in the fecal particulate than that observed with the forages. This interaction also occurred in the determination of acid detergent lignin (ADL). The lower ADL from 72% H_2SO_4 treatment to the ND pre-extracted ADF residue could possibly be due to the use of sodium sulfite in the ND procedure (Van Soest and Robertson, 1979). The further decrease of lignin in Alf particulate digesta obtained from the 72% H_2SO_4 treatment of the ADF-TFA could possibly be due to the combination of ND extraction and TFA hydrolysis. Barton et al., (1982) reported the presence of a lignin-carbohydrate complex (LCC) upon TFA hydrolysis which contained p-coumaric and ferulic-type lignin groups. The lack of consistent extraction by these analyses on fecal particulate are possibly due to the combination of these extractions and in vivo digestion which causes greater DM loss than either treatment alone. Barton et al., (1981) studied the effect of AD reagent and/or incubation with rumen microorganisms in vitro on leaf tissue loss. These authors reported that the response of each grass to the AD reagent varied and that the combination of AD reagent extraction and digestion caused greater tissue loss as shown by scanning electron microscopy (SEM) than either treatment alone. In addition, Windham and Akin (1984) have investigated the effect of these analyses type (i.e., conventional, sequential and TFA hydrolysis) on initial substrate and in vitro indigestible DM remaining (IDMR) of Alfalfa plant parts. An analyses type by plant part (i.e., leaf, stem, and whole plant) interaction was observed in both the initial substrate and in the IDMR. The difference in magnitude between the fiber values obtained were again greater in the in vitro residues than in the initial substrate. Estimates of hemicellulose increased whereas estimates of ADF and lignin decreased with each analyses type (i.e., conventional, sequential, TFA hydrolysis). These data indicate the lack of consistent extraction by the reagents employed even among plant parts of the same forage specie. These analytical data indicate that the combination of digestion followed by extraction by these various methods caused a greater DM loss than either treatment alone.

To further substantiate this loss, free hand sections of Alfalfa stems were examined by SEM for changes in anatomical structure due to the combination of digestion and extraction. The

tissues present in Alfalfa stems are shown in Fig. 1. The secondary xylem, phloem cap, and interfascicular cells are lignified forming a rigid band of thick-walled cells. Whereas, the cortex, vascular cambium, and pith cells are not lignified. Acid detergent reagent removed the unlignified pith and cortex cells and left a residue essentially of the lignified band of cells (i.e., interfascicular and xylem). Sequential treatment of Alfalfa stems did not change the plant structure over that with AD but did result in a slight distortion of the lignified cell walls. Trifluoroacetic acid, however, effected noticeable changes in stem structure with the remaining lignified tissue grossly distorted indicating a loss of cell wall material. After 48 h, rumen microorganisms degraded all but the lignified cells (Fig. 2); at times the cuticle was not always associated with the residue indicating it had sloughed off during incubation. The combination of treatments, (i.e., microbial digestion followed by chemical extraction) resulted in a similar residue of xylem and interfascicular cells (Fig. 3 and 4). However, the remaining tissue was further distorted with total loss of the cuticle. These data and observations indicate that partial destruction of plant fiber in fermentation residues occur by reagents used in the analytical procedures or that physical and (or) chemical differences occur between the initial substrate and fermentation residues such that the nature of the fiber components differ due to the procedures used to empirically define them (Muntifering, 1982).

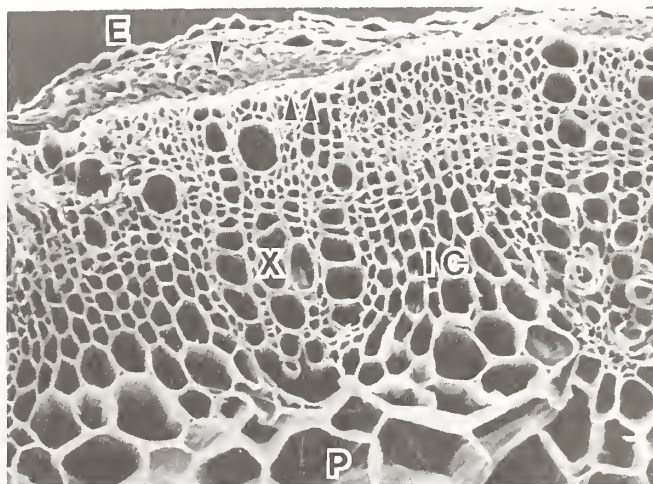


Fig. 1 - Cross section of control Alfalfa stem. Tissues include: epidermis (e), phloem (arrows), vascular cambium (double arrows), xylem (X), interfascicular cells (ic) and pith (P).

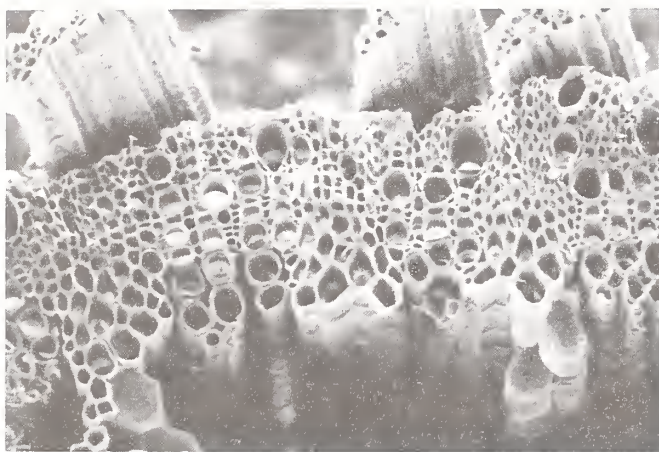


Fig. 2 - Cross section of Alfalfa stem incubated 48 h showing a residue of xylem and interfascicular cells.

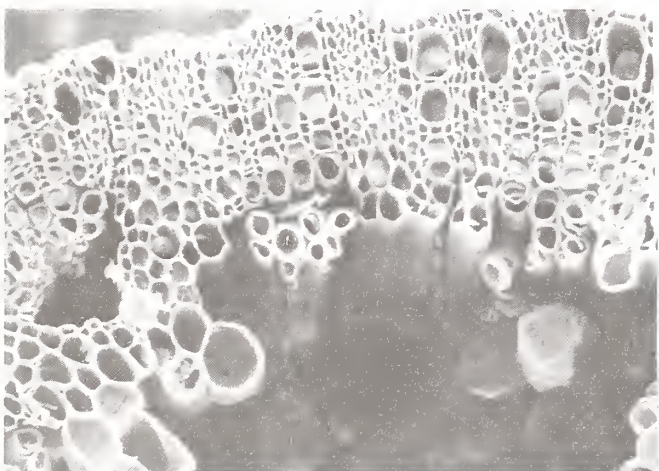


Fig. 3 - Cross section of alfalfa stem incubated 48 h and then treated with AD.

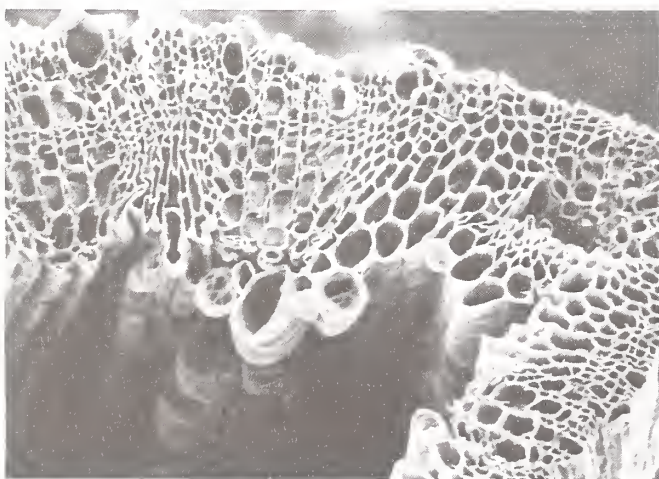


Fig. 4 - Cross section of alfalfa stem incubated 48 h and then treated with ND and TFA.

The major implications of these results are in the interpretation of digesta studies that attempt to determine apparent digestibility coefficients for fiber components. The choice of analytical method may drastically affect the extent of recovery of fiber components and, in turn, digestibility coefficients. In addition, the lack of consistent extraction across forages and fermentation residues by methods illustrates the difficulty of designing a single system of analysis for all conditions.

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C. P. Bagley¹

INTRODUCTION

The Southern Region of the United States has been highlighted as an area with great potential for producing beef animals primarily on forages. Many Experiment Station Directors have labeled the "Forage-Beef" research area as high priority. This is predicated on the ability of the South to grow large amounts of forage using relatively low inputs. The climate of the South is typified as having mild winters, warm summers with rainfall averages between 100 and 150 cm annually. Forages can be grazed 9 to 12 months each year, depending on the forage system employed. The Southern Region has long been heavily involved with beef cow-calf enterprises, having approximately 50% of all the mature cows in the United States located in this region. Most of the calves not kept as replacement heifers are sold at weaning and shipped to other parts of the country for stocker or finishing programs. Much research data has indicated the high efficiency of steers and heifers taken to heavier weights on forages. Many forage sources in the South are not suitable for fattening cattle, but can be used for growing cattle. Taking these animals to heavier weights on forages would conserve grain sources and lessen the public outcry regarding feeding grain to cattle while people are starving.

Therefore, the objective of this manuscript is to outline research work being conducted at the Rosepine Research Station, a branch of the Louisiana Agricultural Experiment Station.

RESEARCH ACTIVITIES

The Louisiana Crop and Livestock Reporting Service estimated that in 1983 there were 648,000 head of beef cows, 110,000 head of replacement heifers and 43,000 head of stocker-finishing steers in Louisiana. The primary goal of the Rosepine Research Station, therefore, is to conduct studies that evaluate systems of beef cattle production using forages as the primary nutrient source. The research involves systems of management for cow-calf production, stocker-finishing operations and raising replacement heifers. Total vertical integration of the beef enterprises is the goal of this research where beef is taken from conception to consumption. Each segment is accounted for in terms of physical inputs and outputs which allows for economic evaluation. In this manner, the relative efficiency of each segment can be evaluated.

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Cow-Calf

A series of studies were begun in 1972 to evaluate systems of management for cow-calf production using forages as the primary nutrient source. The initial study in this sequence involved stocking rates of approximately 2.5 cows per ha using four outcome groups. Pasture systems differed on the availability of creep or forward grazing for calves and the amount of cultivated land in a system used for double cropping summer and winter annuals. Results of this study showed that making available a creep grazing area for calves increased weaning weights by 30 kg. Increasing cultivated land area initially increased calf performance and cow productivity, but cultivating more than 34% of the land resulted in poorer cow performance and much higher cost per unit of gain by calves. The system deemed most advantageous in terms of optimizing calf production while minimizing costs and risk contained 17% cultivated land with creep grazing for calves.

The second study in this series on cow-calf management systems used a single pasture system containing 23% cultivated land planted to winter annuals. The remainder of the pasture system was permanent bermudagrass overseeded annually with ryegrass and white clover. Treatments for cow-calf pairs included comparing fall and spring calving, and stocking rates of 2.9 or 3.7 animal units per ha. Fall calving resulted in heavier calves at weaning, more calf production per hectare, but more supplemental winter feeding. Stocking rates of 2.9 cow-calf pairs/ha resulted in less beef production/ha, but heavier individual weaning weights for the calves, heavier weights for cows, and fewer days on supplemental feed compared to stocking rates of 3.7 animals/ha.

The third and present study being conducted in the series of cow-calf studies involves fall and spring calving using similar stocking rates, with and without cultivated land, with all systems using creep grazing for calves. Cultivated land planted to winter annuals appears promising for increasing weaning weights of calves born in the fall, but spring calving systems do not respond well to cultivated land in the system. This is primarily due to the time of year of this land is out of production.

Stocker-Finishing

Presently, there are six research stations in Louisiana involved with a study entitled "Year Round Production of Finished Beef Using Optimum Levels of Forages". This is part of the Southern Regional cooperative project S-167 of the same title. The project is designed to produce stocker steers (318 kg final weight) at two month intervals throughout the year. Forages are used as the primary nutrient source. One-half of the finished steers are produced on optimum levels of forages while the remaining animals receive a corn silage ration fed at a central feedlot. The corn silage ration is

formulated to contain 57% corn grain and 43% roughage.

Results thus far have indicated that steers can be finished on primarily all forage diets and can be produced at all times of the year while keeping average age of the cattle under 24 months. Minimum target weights have been met at each date. Relative differences do exist between locations, caused by breed types and soil characteristics. In general, input costs are greater for winter pastures on non-alluvial soils compared to alluvial soils, but more grazing days and higher stocking rates can be used due to fewer problems with mud and boggy field conditions. Finishing systems with small grain-ryegrass-clover mixtures have resulted in gains in excess of 1.0 kg per day during the finishing phase. Cost/kg of gain has often been under \$.80 for steers grazed on winter annuals. Costs have been more for steers finished on summer pastures, either perennials or annuals, primarily due to the lower rates of gain.

Numerous combinations and types of summer grazing schemes have been evaluated in this stocker-finishing beef study. Bermudagrass has been successfully used when steers are growing, but gains are poor for heavier animals that are in a finishing phase. Bermudagrass-white clover mixtures have produced acceptable gains for finishing steers. This grass-legume mixture has generally been used during the period between the end of ryegrass grazing and before summer annual grazing is available, due to double cropping. This time period is usually from early June to mid July. Close grazing of the bermudagrass-white clover pasture will allow the white clover to remain in the mixture in sufficient quantities to positively affect animal performance.

Summer annuals used to double crop with winter annuals include millets, alyce clover and sorghum-sudangrass. Tifleaf 1 pearl millet has been successfully grazed for several years. Animal performance has been approximately .6 kg/da with a stocking rate of 12 animals/ha. Alyce clover has yielded gains of almost .8 kg/da for stocker steers, with stocking rates of 5 animals per ha. The sorghum-sudangrass was used only one year, being inferior to millet for both animal performance and stocking rate.

Steers fed corn silage during the finishing phase gained at faster rates, were heavier at slaughter, had higher dressing percentages, and higher carcass quality grades compared to steers finished on all forage diets. However, cost/kg of gain during the finishing phase was much lower for forage-fed steers. Grades of forage fed steers were acceptable with 70% of the steers grading good and choice, compared to 97% grading good and choice when finished on corn silage.

Replacement Heifers

Studies are being conducted which compare breeding heifers to calve initially at either 24 or 30 months of age. Using both spring and fall calving systems allows the advantage of 30 month old calving. Most cow management systems must calve at either 2 or 3 years of age initially due to one season of calving. Heifers bred to calve at 30 months have had slightly higher conception rates initially, and higher conception rates when being bred for second calving compared to heifers calving initially at 24 months.

SUMMARY

The research work at the Rosepine Research Station is involved with numerous aspects of the beef production industry. Because Louisiana and the Southern Region of the United States are primarily involved with cow-calf systems of management, the largest amounts of resources at this station are dedicated to this area of research. Studies at this Station have shown that cow-calf production can be a viable operation if proper management is applied. The stocker-finishing beef production systems have not been exploited in the Southeast as most of these calves are sold at weaning. At present, the stocker-finishing beef system is the only beef enterprise capable of being profitable under present economic conditions. Winter annuals can produce high rates of gain inexpensively. Certain summer forage systems are capable of producing good rates of gain, dependent upon the condition and weight of the cattle.

The cow-calf system is dependent upon the raising of good quality replacement heifers. Age at first calving does affect weaning weight of the calf and the ability of the cow to rebreed. These factors must be evaluated in making choices for breeding heifers.

BUSINESS MEETING AND RELATED MATTERS

Fortieth Southern Pasture and Forage Crop
Improvement Conference
Baton Rouge, LA 70803
19 April 1984
Minutes of the Business Meeting

Dr. Harold Brown, Chairman, called the meeting
to order at 11:30 a.m.

The first order of business was to stand and
recognize the death of Arthur Spooner,
University of Arkansas, by a moment of
silence.

A financial report was made by R. S.
Kalmbacher who indicated that a balance of
\$1,438.85 was present in the SPFCIC account at
Wauchula State Bank.

A motion was made by Dave Timothy, seconded by
Coleman Ward to accept the minutes of the 39th
SPFCIC as prepared and mailed in January 1984.
Passed.

R. S. Kalmbacher outlined the requirements
that SPFCIC must meet in order to receive
tax-exempt status with the U.S. Internal
Revenue Service. The original articles or
"by-laws" from the first meeting in 1940 were
reviewed and a new statement based on approved
changes outlined in the minutes of SPFCIC
(1940 to 1984) was read to the group. It was
pointed out that the only change suggested was
the deletion of the rule that "associated work
groups will coordinate their programs with the
host". In its place the statement "The past
chairman will have the responsibility for the
general session program and work groups
coordination." The role of Secretary/
Treasurer was outlined and also instructions
were provided for dispersant of SPFCIC funds
should SPFCIC abandon its function. Copy of
this was article is attached. A motion was
made by Doug Chamblee, seconded by J. P.
Muller to accept the document as read.
Passed.

Dr. Doug Chamblee read a resolution honoring
Dr. Timothy Taylor and recognizing his
contributions to SPFCIC. Dr. Chamblee made a
motion, seconded by Warren Thompson, to have
the 40th SPFCIC adopt this resolution.
Passed. The Secretary/Treasurer was
instructed to send this resolution to Dr.
Taylor. A copy of this resolution is
attached.

Dr. Dave Timothy outlined the 1985 SPFCIC
meeting in Raleigh, NC. The dates of this
meeting will be 20 May (evening), 21 and 22
May 1985.

Dr. Harold Brown read a brief letter of
invitation from Dean W. P. Flatt of the
University of Georgia. Dr. Brown outlined
plans for the 1986 meeting in Athens, Georgia,
and proposed that the SPFCIC meeting be held
in the same week (14 April 1986) as the annual

AFGC meeting with a joint tour. A motion was
made by Carl Hoveland, seconded by Harlan White
to hold a joint SPFCIC-AFGC meeting in April
1986. Passed.

A report of the nominating committee was made by
Dr. Marvin Riewe. The committee consisted of
Carl Hoveland and Joe Burns (Tennessee). Hagen
Lipke was nominated for chairman, elect-elect.
Dr. Brown asked for nominations from the floor,
and hearing none, stated that Dr. Lipke was
elected by acclamation.

Dr. Coleman Ward, chairman of the resolutions
committee, which consisted of Harry Bryant and
Vance Watson, read a resolution recognizing the
host institution and all people who made the
40th meeting possible. A copy of this
resolution is attached. A motion was made by
Joe Fontenot, seconded by Joe Burns to adopt the
resolution. Passed.

Dr. Brown, having completed his duties as
chairman, passed the gavel and responsibilities
of chairman to Dr. Billy Nelson.

Billy Nelson, chairman, presented Dr. Brown with
a plaque recognizing his contributions to
SPFCIC.

A motion was made to adjourn. Passed.

Resolution 1.

WHEREAS, Timothy H. Taylor has participated
actively in the Southern Pasture and Forage Crop
Improvement Conference for more than 28 years
serving as Chairman in 1977, and

WHEREAS, he has served with distinction on
team research efforts to solve many of the
ecological problems of grassland production and,

WHEREAS, he has developed biological and
mechanical principles and practices designed to
introduce legumes into fields of cool-season
grass sods; and pioneered in the study of the
development, senescence, and photosynthetic
activity of orchardgrass leaves; and developed
establishment and cutting management systems for
many of our major forage species.

BE IT THEREFORE RESOLVED that we recognize
our friend and professional colleague, DR.
TIMOTHY TAYLOR, and thank him for his many
contributions to the improvement of grassland in
our region, and wish him many years of happiness
during his retirement period.

SOUTHERN PASTURE AND FORAGE CROP IMPROVEMENT CONFERENCE

Resolution 2.

WHEREAS, the membership of the 40th Annual
Southern Pasture and Forage Crop Improvement
Conference has gleaned much information and
great benefits from its participation in the

conference, and

WHEREAS, such information and benefits could not have been realized without the friendly, hospitable and concerted efforts of the staff and administration of Louisiana State University, it's Agricultural Center and the Louisiana Agricultural Experiment Station.

BE IT RESOLVED that the 40th Conference express its grateful appreciation to the staff, faculty and administration of Louisiana State University for their gracious hospitality, imaginative programming, well planned and executed tour of the forage and livestock industry of Southeastern Louisiana, which was of great interest to the membership, and especially for their outstanding purposeful research to obtain answers to underlying causes and basic problems of forage and livestock agriculture.

THAT special recognition be extended to H. Rouse Caffey, Chancellor LSU Agricultural Center, Doyle Chambers, Vice Chancellor for Research and Director of the Louisiana Agricultural Experiment Station, K. W. Tipton, Associate Director, LAES and Denver T. Loupe, Vice Chancellor and Director, Louisiana Cooperative Extension Service.

SIGNAL RECOGNITION be extended to the individuals who served on the local arrangements committee:

- Billy Nelson - General & Program Chairman
- Ann Marie Thro - treasurer
- Ernest Morgan and Cliff Mondart - tour coordinators
- Nelson Philpot and Lee Mason - finances
- Dan Robinson - facilities and housing
- Allan Nipper and Charles Montgomery - audio-visual
- Ann Marie Thro, Wade Faw, and Diana Friesner - registration
- Pat Bagley and Marvin Allen - banquet and entertainment

TO TOUR HOSTS - Johnny Green, President and Dwight Jackson, Manager, Haywood Green Farm; Kirby and Marsha Varnado of V-Clearview Holstein Farm; Lee Mason, Resident Director; the staff and faculty of the Southeast Research Station, Franklinton, LA for the tour of their research and laboratories and for the excellent luncheon.

TO: Floyd Kent, banquet speaker

TO: Conference Chairman, R. Harold Brown; Immediate Past CHairman (and Program Chairman), Harlan White; and Secretary, Robert Kalmbacher.

TO: Session Chairman, Monte Rouquette, physiology-ecology; B. V. Conger, breeding; David Mertens, utilization; and Loren Ramman, extension.

TO: All who presented conference papers

TO: Robert Godke for his discussion on embryo transfer

ADDITIONAL RECOGNITION is given to the following firms who contributed financially to the conference:

Alexandria Seed Company, Alexandria, LA
Citrus Lands of Louisiana, Belle Chase, LA
Dairy, Inc., Franklinton, LA
KeKalb-Pfizer Genetics, Monroe, LA
Freeport Mc-Mo-Ran, Inc., New Orleans, LA
Funk Seeds International, Alexandria, LA
Louisiana Forage and Grassland Council, Baton Rouge, LA
Travel Sales Corporation, Folsom, LA
Terral-Norris Seed Co., Lake Providence, LA

The Resolutions Committee
Harry T. Bryant
Vanch H. Watson
Coleman Y. Ward, (Chairman)

Organization and By-Laws of the Southern Pasture and Forage Crop Improvement Conference

History: At the annual meeting of the American Society of Agronomy in New Orleans, December 1939, there was informal discussion regarding the desirability of pasture and forage crop specialists in the South holding a conference to discuss mutual problems common to the Southern states. Following the Agronomy meetings, the procedure for organization was discussed with experiment station officials and pasture specialists in various states. The general opinion seemed to be that the most representative organization in the South to sponsor such a conference group would be the Association of Southern Agricultural Workers. Mr. R. L. Lovvorn of the North Carolina Experiment Station called a conference of research workers interested in pasture and forage crops at the annual Southern Agricultural Workers meeting in Birmingham, Alabama on 7 February 1940.

At this first organization meeting the temporary chairman appointed a committee to formulate plans for organization. The committee was instructed to report at a second meeting of the group on 8 February 1940. The committee was as follows:

G. W. Burton, Georgia
D. G. Sturkie, Alabama
G. E. Ritchey, Florida
H. W. Bennett, Mississippi
R. L. Lovvorn, North Carolina

At the second organizatin meeting these following recommendations for organization were adopted:

Name: The name shall be 'The Southern Pasture and Forage Crop Improvement Conference'.

Membership: Membership shall consist of workers actively engaged in pasture and forage crop improvement.

assets in the treasury will be donated to the American Forage and Grassland Council (tax-exempt No. 25-1155822).

Objectives:

1. To encourage the cooperation of all groups that are in a position to contribute to the pasture and forage crop improvement program.
2. To aid in the coordination of activities between workers.
3. To assist in the development of the program of the Association of Southern Agricultural Workers.
4. To act as a medium of exchange of ideas, methods of procedure and needs of workers.
5. To act as an agency for the exchange of plant material.
6. To assist in the formation of a policy for the distribution of material.

Officers: The conference shall be guided by an Executive Committee of nine members. Four members (officers) shall be elected for a term of four years, during which time they shall bear the title: Chairman elect-elect, Elect, Immediate, and Past-chairman, respectively. It is suggested that a nomination committee, appointed by the present chairman, select one candidate annually to be confirmed as chairman elect-elect. The office of Secretary-Treasurer will be the fifth member elected for a term of three years. The remaining four members shall be representatives of each of the four interest groups: Forage Breeding, Forage Ecology and Physiology, Forage Utilization, and Forage Extension. Each interest group will have one vote.

Meetings: The meeting of the SPFCIC will be held annually. A university administration wishing to host a meeting will make a formal invitation to the chairman two years prior to the meeting date. The order established is as follows: Virginia, Oklahoma, Louisiana, North Carolina, Georgia, South Carolina, Kentucky, Arkansas, Texas, Mississippi, Alabama, Florida, and Tennessee. The host institution will provide for local arrangements, field trips and other activities at their discretion. The past chairman will have the responsibility for the general-session program and work-group coordination.

Treasury: The Secretary will function as Treasurer of the Southern Pasture and Forage Crop Improvement Conference and will be accountable for receipt and disbursement of money. Receipts from registration or from industry contributions that are not spent for conduct of the meeting of the current year will be maintained for the organization of the meeting in the subsequent year. These cash reserves will be used for printing, postage, and other operational expenses. If the Southern Pasture and Forage Crop Improvement Conference is permanently dissolved, the

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